Journal of the Fisheries Research Board of Canada

Journal de l'office des recherches sur les pêcheries du Canada

Volume 32, No. 3, March 1975

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Environment Canada
Fisheries and Marine Service

Environnement Canada
Service des pêches et des sciences de la mer

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Office of the Editor
116 Lisgar, Ottawa K1A 0H3

Bureau du Rédacteur

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Journal of the Fisheries Research Board of Canada

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Biodegradation of Di-*n*-Butyl Phthalate and Di-2-Ethylhexyl Phthalate in Freshwater Hydrosoil[1]

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Johnson, B. T., and W. Lulves. 1975. Biodegradation of di-*n*-butyl phthalate and di-2-ethylhexyl phthalate in freshwater hydrosoil. J. Fish. Res. Board Can. 32: 333–339.

The phthalic acid esters di-2-ethylhexyl phthalate (DEHP) and di-*n*-butyl phthalate (DBP) which are used as plasticizers and recovered in routine chemical analysis of freshwater fish, were incorporated into freshwater hydrosoil in the laboratory. Samples containing about 1 mg/liter of these two esters, ^{14}C (carbonyl) labelled, were incubated aerobically and anaerobically for 1, 5, 7, 14, and 30 days. Differences in the rates and conditions of degradation of the two esters were marked. Under aerobiosis, 53% of the radio-labelled DBP was degraded within 24 h, and 98% within 5 days; DEHP, in contrast, was only 50% degraded after 14 days. Under anaerobiosis degradation of both esters was retarded. DBP was degraded only one-sixth as fast in hydrosoil overlayed with nitrogen whereas degradation of DEHP was not detected. Our evidence from both thin-layer chromatography and radiorespirometry suggests that the esters undergo decarboxylation after initial hydrolysis, probably to the 1,2-dihydroxybenzene molecule.

Johnson, B. T., and W. Lulves. 1975. Biodegradation of di-*n*-butyl phthalate and di-2-ethylhexyl phthalate in freshwater hydrosoil. J. Fish. Res. Board Can. 32: 333–339.

Les esters di-2-éthylhéxyl phthalate (DEHP) et di-*n*-butylphthalate (DBP) de l'acide phthalique, utilisés comme plastifiants et récupérés lors d'analyses chimiques routinières de poissons d'eau douce, ont été incorporés à un hydrosol d'eau douce au laboratoire. Des échantillons contenant environ 1 mg/litre de ces deux esters, marqués au ^{14}C (carbonyl), ont été incubés dans des conditions aérobies et anaérobies durant 1, 5, 7, 14 et 30 jours. La vitesse et les conditions de dégradation des deux esters accusent des différences marquées. Dans des conditions aérobies, 53% du DBP marqué est dégradé en dedans de 24 h et 98% en dedans de 5 jours; par contre, DEHP n'est dégradé que de 50% au bout de 14 jours. Dans des conditions anaérobies, la dégradation des deux esters est retardée. Dans un hydrosol enduit d'azote, la dégradation du DBP

[1]Trade names referred to in this paper do not imply endorsement of commercial products.

Printed in Canada (J3569)
Imprimé au Canada (J3569)

se produit à un rythme ralenti d'un sixième, alors que nous n'avons pas détecté de dégradation de DEHP. La chromatographie en couche mince, de même que la radiorespiratométrie, suggèrent que les esters, après hydrolyse initiale, subissent une décarboxylation, probablement vers la molécule 1,2-dihydroxybenzène.

Received September 16, 1974
Accepted December 2, 1974

Reçu le 16 septembre 1974
Accepté le 2 décembre 1974

REPORTS of phthalic acid esters (PAEs) in soil (Ogner and Schnitzer 1970), water (Corcoran 1973; Hites 1973), and fish (Mayer et al. 1972) have appeared in the recent literature. Mathur (1974) has only recently reviewed the topic of phthalate esters in the environment. During 1972, an estimated 1 billion pounds of 20 different phthalates were manufactured in the United States (Hall 1971). Although PAEs, the esters of benzene ortho dicarboxylic acid, have a broad spectrum of uses ranging from defoaming agents to perfume vehicles in cosmetic production (U.S. Tariff Commission 1971), they are utilized chiefly as plasticizers with the poly (vinyl chloride) polymers (PVCs). Used in conjunction with PVCs, plasticizers impart flexibility, workability, and extensibility to the product (Graham 1973). In as much as PAE plasticizers have a many faceted market in automotive, construction, clothing, home furnishing, medical, and packaging industries (U.S. Tariff Commission 1971), their appearance in aquatic organisms and habitats seems inevitable. Even though PAE residues are apparently ubiquitous in the environment, the acute and chronic effect on man and wildlife remains essentially unknown — "an etiology in search of a disease" (Tepper 1973). Fragmentary evidence suggests potential deleterious effects on freshwater biota. Sanders et al. (1973) and Mayer and Sanders (1972) reported reproductive inhibition and bioaccumulation in the filter-feeding cladoceran *Daphnia magna*. Metcalf et al. (1973) also found rapid accumulation of phthalates in a variety of plants and animals in a model ecosystem. The fate of PAEs in the aquatic environment remains largely a mystery. Saeger and Tucker (1973) demonstrated that all PAEs tested underwent complete aerobic degradation, at rates comparable with those of a readily degradable linear alkyl benzene sulfonate, in the presence of activated sewage sludge and river water. Unfortunately, no data are available concerning degradation under the anaerobic conditions that frequently attend hydrosoils in the natural environment. A number of questions concerning the ultimate fate of PAEs remain unanswered. Do PAEs rapidly undergo complete biodegradation in the aquatic environment to harmless elemental fragments or are they resistant to microbial attack? Are the current PAE residues a reflection of persistency or an equilibrium between input and natural decomposer activity?

In this investigation, we report on the biodegradation of two widely used plasticizers, di-2-ethylhexyl phthalate (DEHP) and di-*n*-butyl phthalate (DBP) by unidentified aerobic and anaerobic microorganisms found in hydrosoil of freshwater ponds. The studies were performed in the laboratory under aerobic and anaerobic conditions, with ^{14}C-labelled phthalic acid esters. The microbial degradation of phthalates was determined by thin-layer chromatography–autoradiography and measurement of $^{14}CO_2$ released from labelled phthalates.

Materials and Methods

CHEMICALS

^{14}C-carbonyl-labelled di-*n*-butyl phthalate (1.53 mCi/mM), di-2-ethylhexyl phthalate (1.64 mCi/mM) and phthalic acid (2.74 mCi/mM) were purchased from Mallinckrodt Co. Standards of DBP and DEHP were obtained from Monsanto Company. Chemicals listed in Table 4 were purchased from Chem Service or Eastman Kodak Chemical Co. The ^{14}C-*n*-butyl phthalate (monoester, BP) was synthesized from ^{14}C-DBP by *Bacillus subtilis* (Fish-Pesticide Research Laboratory Collection). Acetone was used as the solvent carrier for all compounds investigated (<0.01% vol/vol). Plate counts were made of control samples containing 0.1% (vol/vol) redistilled, analytical grade acetone at 1-, 7-, and 30-day intervals. We found no evidence that acetone influenced the viability of the microflora in hydrosoil or induced any radical pH changes in the cultures during incubation.

CULTURING AND EXTRACTION METHODS

Hydrosoil samples were taken from 0.04- and 0.10-ha freshwater ponds located at the Fish-Pesticide Research Laboratory. We used a core-type sampling device (Holz et al. 1972) to collect pond bottom material along the shoreline at a water depth of about 1 m. We removed the hydrosoil to a depth of 5 cm in the core sample at the mud–water interface and screened it through a 40-mesh wire sieve to eliminate macroinvertebrates. Plate counts of the hydrosoil, incubated both aerobically and anaerobically at 22 C, showed an average population of 5×10^6 viable organisms per milliliter on brain–heart infusion agar

(BBL). All hydrosoil samples contained 5% organic carbon (dry weight) (American Public Health Association 1971). Ten grams (wet weight) of hydrosoil were placed into a 50-ml Delong flask and covered with 20 ml of pond water. The flask content was then inoculated with 100 μl of acetone containing the ^{14}C-labelled phthalate. Cultures were adjusted to contain about 1 mg/liter labelled phthalate. After inoculation, the aerobic groups were incubated in a water bath rotary shaker (180 rpm, 1 in orbit) and the anaerobic groups were placed in an anaerobic jar (Torsion Co.), evacuated under vacuum, and overlayed with nitrogen. All samples were incubated at 22 C. We tested for spontaneous degradation (abiotic) of the phthalates by including an autoclave-killed control. The control sample was autoclaved at 15 lb pressure and 121 C for 20 min. We added 250 mg/liter sodium azide to some hydrosoil samples as a chemical means of inhibiting microbial activity (Kaufman et al. 1968). Hydrosoil was maintained under aerobic or anaerobic conditions and sampled in triplicate at 1-, 5-, 7-, and 30-day intervals. Controls consisted of acetone and autoclave-killed hydrosoil. All samples were extracted on the day of collection. Each hydrosoil sample was initially acidified to pH 1-2 with concentrated H_3PO_4 and extracted 3 times with an equal volume of diethyl ether. The ether extracts were centrifuged for 10 min at 10,000 × g to remove particulate matter and break emulsions, combined, dried over anhydrous Na_2SO_4, and carefully evaporated to about 1 ml at room temperature under a hood. Extraction efficiency of the phthalates from hydrosoil was routinely 85 ± 5%; all data are based on this accuracy value. Autoradiography of thin-layer chromatograms indicate both ^{14}C–DBP and ^{14}C–DEHP were 99+% pure.

Chromatography–Autoradiography

The ether extract was spotted by means of a Camag Linomat® automatic TLC spotter on 0.2-mm precoated silica gel thin-layer chromatography plates (Brinkman, EM Reagents, Silica Gel, F-254) containing a fluorescent indicator. The TLC plates were developed 3 times in petroleum ether:diethyl ether:acetic acid (77:20:3) (vol/vol/vol) (Stalling et al. 1973). We used no-screen medical X-ray film to visualize radioactive products by autoradiography and shortwave ultraviolet light (2537Å) to detect non-labelled standards and controls. The quantity of each radio-labelled material separated on the chromatogram was determined by scraping silica gel from the specific radioactive R_f area into a scintillation vial containing 15 ml of toluene:fluor (Fluoralloy®, Beckman Instrument Co.) and then counting the mixture with a Beckman 200-L liquid scintillation counter set for ^{14}C-β-particle energy spectrum. Results were corrected for quench and background radiation.

Radiospirometry

We constructed a radiospirometer to detect possible $^{14}CO_2$ evolution from the ^{14}C-phthalate hydrosoil sample during incubation. Hydrosoils containing the two ^{14}C-diesters were sampled at 7-, 14-, 21-, and 28-day intervals for $^{14}CO_2$ production. Controls consisted of both autoclave-killed and NaN_3 samples. Air was passed at a flow rate of 5 ml/min into the experimental chamber after being scrubbed with 2 N NaOH to remove CO_2. The effluent gas from the incubation chamber was first passed through a 50.4- × 6.35-mm column containing OV-225 coated Chromosorb W HP and finally bubbled through a monoethanol amine:ethylene glycol-CO_2 (3:7 vol/vol) trapping solution. The $^{14}CO_2$ was trapped as a carbamate. To investigate the efficacy of this system, we evaporated ^{14}C-labelled phthalic acid, DBP, DEHP, and BP in a small flask, connected it to the respirometer, and gently warmed the flask to vaporize the materials. We found only trace amounts of radioactivity in the CO_2-trapping solution. The major portion of the radioactivity recovered was confined to the Chromosorb W column. Triplicate, 1-ml samples of the $^{14}CO_2$-mono-ethanol amine solution were added to 10 ml of a dioxane-fluor mixture composed of 4 g of 2,5-diphenyloxazole (PPO), 200 mg of 1,4 *bis*-2-(4-methyl-5-phenyloxyazolyl)-benzene (POPOP), 100 ml of methanol, and 60 g of naphthalene in 1 liter of dioxane. Scintillation samples were counted with the Beckman 200-L liquid scintillation system and corrected for quench and background radiation.

Statistical Inference

Mean comparisons were made according to Fisher's "*t*" test; significance was taken at $P < 0.05$. All data represent the mean value of samples removed from triplicate incubation vessels ($\bar{x} \pm SE$).

Results and Discussion

Freshwater hydrosoil degraded both ^{14}C-DBP and ^{14}C-DEHP (Tables 1, 2, 3). Since autoclave-killed or NaN_3 controls did not significantly degrade either ester, we attributed the degradation of the two phalates to the enzymic action of microorganisms indigenous to freshwater hydrosoil.

We found marked differences between the conditions and rates of biodegradation of the two esters. Forty-Six percent of the radio-labelled DBP in hydrosoil (Table 2) was degraded aerobically to mono-*n*-butyl phthalate within 24 h and nearly 98% (Table 1) of all radioactivity disappeared after 5 days. Under the same conditions, nearly 14 days were required for 53% of the DEHP-labelled material to disappear (Table 2).

Anaerobiosis slowed biodegradation of both esters. Although nearly 98% of the radio-labelled DBP disappeared during 30 days of incubation in the hydrosoil, the retarding influence of anaerobiosis was evident. Nearly 70% of the DBP radioactivity remained after 5 days of incubation

TABLE 1. Biodegradation of ^{14}C-di-*n*-butyl phthalate and ^{14}C-di-2-ethylhexyl phthalate in freshwater hydrosoil.

Incubation (*days*)	Percent recovery of radioactivity from hydrosoil	
	Aerobic	Anaerobic
^{14}C-di-n-butyl phthalate		
1	95[b]	100[b]
5	3[c]	69[c]
7	5[c]	59[c]
14	8[c]	39[c]
30	3[c]	2[c]
Heat-killed control[d]	100	100
^{14}C-di-2-ethylhexyl phthalate[ac]		
7	100[b]	100[b]
14	53[c]	100[b]
30	41[c]	100[b]
Heat-killed control[d]	100	100

[a]Data represent the mean value of triplicate samples. Precision = 85 ± 5%. Values rounded to nearest 1.0.

[b]No significant difference existed between control ($P < 0.05$, Fisher's "*t*").

[c]Significant difference between control ($P < 0.05$, Fisher's "*t*").

[d]30-day incubation.

[e]Degradation products were 2% of total radioactivity recovered.

under anaerobic conditions, whereas only 3% of the extractable labelled DBP and degradation products remained after 5 days of aerobic incubation. Degradation in anaerobic hydrosoils did not reach the aerobic level until the 30-day sampling. Ester hydrolysis required twice as much time in anaerobic as in aerobic hydrosoil, and decarboxylation time was about 6 times longer under anaerobic conditions. DEHP was completely resistant to microbial attack under anaerobic conditions. After 30 days, we observed no significant loss of ^{14}C-DEHP activity in hydrosoils overlayed with nitrogen (Table 1).

IDENTIFICATION OF DEGRADATION PRODUCTS

In TLC-autoradiography analyses of the ^{14}C-DBP and ^{14}C-DEHP hydrosoil extracts we repeatedly found radioactivity in DBP extracts at R_f 0.82, 0.33, 0.25, 0.15, 0.09, and 0.00 (Table 2). The spots at R_f 0.82, 0.33, and 0.09 (Table 4), when cochromatographed with nonradio-labelled authentic standards, were similar to the spots of di-*n*-butyl phthalate, *n*-butyl phthalate, and phthalic acid, respectively. The spots at R_f 0.25, 0.15, and 0.00 did not correspond to known degradation products (Table 4). Quantitation of the radioactivity at each R_f value by liquid scintillation spectrometry indicated that the major degradation product of the 1-day sample was the monoester (46.3%). Small amounts of compounds more polar than the monoester (6% of the total recovered radioactivity) were at R_f 0.25, 0.09, and 0.00. R_f 0.09 corresponded to phthalic acid; R_f 0.25 and 0.00 were unidentified compounds.

Significantly, recovery of radioactive material showed that samples incubated for 1 day contained 95% of the total radioactivity found in the control. This recovery decreased in the 5-, 7-, 14-, and 30-day samples. In comparison with the control, only 2.9–7.9% of the radioactivity introduced into these samples was recovered (Table 2). Qualitative analyses of these extracts by TLC-autoradiography revealed the parent diester, the monoester, phthalic acid, and a trace amount of an unknown or unknowns at the origin in an approximate ratio of 76:18:3:1% (Table 2).

The slower rate of PAE degradation under anaerobic than under aerobic conditions is clearly reflected in the type and quantities of the radioactive components extracted from hydrosoils after various incubation intervals (Table 1, 2). At least four compounds were detected in anaerobic DBP samples: the monoester, phthalic acid, and two unknowns at R_f 0.25 and 0.00. The compound or compounds at R_f 0.15 was not seen in any of the autoradiograms taken from anaerobic extracts. Although *n*-butyl phthalate was the major degradation product in either the aerobic or anaerobic samples after 1 day of incubation, the degradation rate was more than 1.5 times faster under aerobic conditions (32% vs. 50%). Qualitative comparison of aerobic with anaerobic phthalate degradation after the first 24-h incubation period is at best tenuous because of the rapid loss of radioactivity from the samples. We did, however, observe an increase in the monoester after long incubation and an apparent lack of accumulation of the more polar degradation products in anaerobic samples (Table 2).

TLC-autoradiographic analysis of aerobic DEHP extracts revealed radioactive spots at R_f 0.85 and 0.36. Cochromatographic data with authentic standards suggest that the products were di-2-ethylhexyl phthalate and the monoester, 2-ethylhexyl phthalate. Liquid scintillation quantitation showed only 2% of the total recovered radioactivity as the monoester. We found no other compound in the aerobic extracts. After 30 days, hydrosoils incubated anaerobically showed no detectable DEHP degradation.

TABLE 2. Effect of incubation on the recovery of di-*n*-butyl phthalate from a freshwater hydrosoil.

Radioactive compound	R_f value[b]	% Recovered radioactive compound after day(s) incubation[a] 1	5	7	14	30
Aerobic Incubation						
Di-*n*-butyl phthalate	0.82	46.3±5.7	85.3±2.8	70.0±2.1	74.0±2.1	76.0±6.5
Mono-*n*-butyl phthalate	0.33	46.3±5.0	7.0±2.1	26.0±2.0	22.0±1.5	18.6±6.1
Unknown I	0.25	2.3±0.3	Trace[d]	0	0	0
Unknown II	0.15	0.1±0.01	0	0	0	0
Phthalic acid	0.09	0.9±0.06	2.3±1.2	3.0±0.5	2.3±0.6	3.6±1.3
Origin	0.00	2.3±0.05	3.3±0.3	1.0±0.05	1.0±0.01	1.0±0.01
% Recovery DBP compared to control[c] at day(s)		95.0	2.9	5.4	7.9	2.7
Anaerobic Incubation						
Di-*n*-butyl phthalate	0.82	68.3±10.0	29.6±8.9	29.0±8.1	5.0±1.5	37.6±12.1
Mono-*n*-butyl phthalate	0.33	30.7± 5.0	45.3±4.5	63.0±9.0	85.6±5.0	16.9± 6.5
Unknown I	0.25	T[d]	4.3±0.3	1.3±0.3	1.0±0.03	0
Phthalic acid	0.09	0.5± 0.01	16.3±4.6	4.6±3.2	6.3±2.7	35.0±11.7
Origin	0.00	T	4.6±1.4	T	T	9.0± 2.5
% Recovery DBP compared to control[c] at day(s)		100.0	69.1	58.5	39.1	2.0

[a]TLC-autoradiographic analysis; data represent mean value of triplicate samples ($\bar{x} = \pm SE$).
[b]Solvent system = petroleum ether:diethyl ether:acetic acid (77:20:3 vol/vol/vol).
[c]Autoclave-killed control:aerobic 158,109 ± 3,165 dpm and anaerobic 166,018 ± 14,758 dpm ($\bar{x} \pm SE$).
[d]Trace = 0.01% total recovered radioactivity.

TABLE 3. Aerobic degradation of ^{14}C-di-*n*-butyl phthalate, ^{14}C-di-2-ethylhexyl phthalate, ^{14}C-mono-*n*-butyl phthalate, and ^{14}C-phthalic acid by freshwater hydrosoil in a radiorespirometer.

^{14}C-labelled compound	$^{14}CO_2$ evolved (total dpm × 10^3)[a]					Total dpm	
	3 days	7 days	14 days	21 days	28 days	Recovered as $^{14}CO_2$	Introduced as ^{14}C-compound[b]
di-*n*-butyl phthalate	ND	24.3	8.6	5.0	1.6	39.2	50.0
di-2-ethylhexyl phthalate	ND	2.1	12.0	8.5	7.6	30.2	50.0
mono-*n*-butyl phthalate	12.4	23.0	ND	ND	ND	35.4	38.0
phthalic acid	97.3	181.1	ND	ND	ND	278.4	268.6
Control[c]	0	0	0	0	0	0	0

[a] $^{14}CO_2$ trapped in monoethanol amine:ethylene glycol. Data represent average of duplicate samples. ND = not done.
[b] Approximate value ± 5.0%.
[c] All heat-killed and NaN_3 controls showed < 0.1% total dpm introduced as $^{14}CO_2$.

DECARBOXYLATION IN HYDROSOIL

We attributed the rapid loss of radioactivity from the hydrosoils to biodegradation of the labelled phthalates during incubation (Table 3). We speculate that the ^{14}C-labelled portion of the phthalate molecule, the carbonyl group, is exposed to further degradation after initial hydrolysis of the ester-linkage. Subsequently, the labelled carbon is removed from the mono-phthalate (or phthalic acid, salicylate, or benzoate) moiety in the form of $^{14}CO_2$ by decarboxylation. To test this hypothesis, we inoculated ^{14}C-*n*-butyl phthalate (BP) and ^{14}C-phthalic acid into hydrosoils and incubated them in a radiorespirometer. During aerobic incubation, we detected no ^{14}C-volatile compounds other than $^{14}CO_2$ escaping from the hydrosoil into the effluent gas and no radioactivity in the toluene extract from the OV-225 column.

Recovery of radioactivity from the CO_2-trapping solution closely paralleled the disappearance of ^{14}C-labelled material from TLC-autoradiograms of both ^{14}C-DBP and ^{14}C-DEHP (Table 3). Only trace amounts of $^{14}CO_2$ activity were recovered from heat-killed and NaN_3 controls. We interpret these data to indicate that the ^{14}C-labelled material in the monoethanol amine solution is carbon-14 labelled carbon dioxide in the form of ^{14}C-carbamate; the evolution of $^{14}CO_2$ is due to the direct enzymic activity of the hydrosoil microflora on the ^{14}C-phthalates.

TABLE 4. R_f values of phthalic acid esters and potential degradation products.

Compound	R_f value[ab]
n-butyl salicylate	0.90
Di-2-ethylhexyl phthalate	0.85
Di-*n*-butyl phthalate	0.81
Benzoic acid	0.60
Salicylic acid	0.51
Mono-2-ethylhexyl phthalate	0.36
Mono-*n*-butyl phthalate	0.33
p-hydroxybenzoic acid	0.20
3-hydroxyphthalic acid	0.16
Phthalic acid	0.09

[a] Solvent system = petroleum ether:diethyl ether:acetic acid, 77:20:3 (vol/vol/vol) TLC plates were developed 3 times.
[b] Values are the mean of five replicates.

PAE DEGRADATION PATHWAY

We propose a pathway for the biodegradation of DBP and DEHP by microflora in freshwater hydrosoil. The proposed scheme is predicated on TLC-autoradiography evidence. Under either aerobic or anaerobic conditions, the diesters are initially hydrolyzed at the ester linkage to form the phthalic acid half-ester and the corresponding alcohol. We believe this monoester is subsequently degraded, primarily to phthalic acid, by continuous esterase activity in the hydrosoil. We suspect that the exposed carboxyl group of the monoester undergoes decarboxylation (either oxidative or reductive) to form the benzene or salicylate derivatives. The R_f values of authentic standards of benzoates and salicylates, however, exceed the R_f values of 0.25 and 0.15 deter-

mined from these unknowns (Table 4). Phthalic acid is rapidly decarboxylated (Table 3), we postulate, to form either the 1,2-dihydroxy benzene (catechol) or benzene. We believe the hydroxylated compound is the most probable product. Meikle (1972) indicated that when the carboxyl group is attached directly to the aromatic ring, the most common microbial attack on the molecule is via the oxidation decarboxylation pathway. Aerobic and anaerobic investigation of ^{14}C-*n*-butyl phthalate and ^{14}C-phthalic acid (both carboxyl-labelled) tend to support this biodegradation pathway (Table 3). TLC-autoradiograms of ^{14}C-BP hydrosoil extracts reveal the disappearance of the monoester concomitant with the appearance of trace amounts of material at R_f 0.09 (phthalic acid) and loss of radioactivity from the TLC plates. Autoradiograms of phthalic acid extracts showed no detectable degradation products, and furthermore, after 7 days incubation the hydrosoil extract contained less than 5% of the radioactivity that was recovered from the autoclave-killed control ether-extract. Radiorespirometry data showed rapid decarboxylation of phthalic acid in hydrosoil (Table 3). For example, we recovered 40.1 and 75.8% of the ^{14}C-phthalic acid radioactivity as trapped $^{14}CO_2$ after incubation in hydrosoil for 3 and 7 days, respectively. Similarly, in hydrosoil, nearly 33% of the radioactivity of the monoester ^{14}C-BP appeared as $^{14}CO_2$ in 3 days and nearly twice that amount in the next 4 days. In general, the disappearance of radioactivity recovered from TLC-plates closely paralleled the recovery of $^{14}CO_2$ trapped in the respirometer samples. No radioactivity was detected in the OV-225 Chromosorb W column from either ^{14}C-phthalic acid or ^{14}C-BP samples.

There is extensive literature on the microbial destruction of the aromatic nucleus, leading to the ultimate utilization of the C-fragment by microorganisms (Evans 1963; Gibson 1968, 1972). Unfortunately, we were unable to obtain substantial evidence to suggest ring cleavage after ester hydrolysis because the phthalic acid moiety was only ^{14}C-carbonyl labelled.

Acknowledgments

We thank J. Johnson, J. Hogan, F. Mayer Jr., R. Schoettger, and D. Stalling for their advice and assistance.

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Population Dynamics of Limnetic Cladocera in a Beaver Pond

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KWIK, J. K., AND J. C. H. CARTER. 1975. Population dynamics of limnetic Cladocera in a beaver pond. J. Fish. Res. Board Can. 32: 341–346.

In a small, shallow, predation free beaver pond near Georgian Bay, Ontario *Ceriodaphnia quadrangula* was monacmic and *Daphnia ambigua* and *Bosmina longirostris* triacmic. Each species peaked and declined rapidly, presumably overshooting the carrying capacity of a food limited environment. Embryonic development times of each species at different temperatures was determined in the laboratory and fitted with Bělahrádek's function. Calculated instantaneous rates of birth and death were normal for *D. ambigua* and *C. quadrangula* but too low to account for the rapid fluctuations in numbers of *B. longirostris*, suggesting occasional gross sampling errors. *Bosmina longirostris* may periodically abandon the limnoplankton for a benthic existence thus avoiding capture.

KWIK, J. K., AND J. C. H. CARTER. 1975. Population dynamics of limnetic Cladocera in a beaver pond. J. Fish. Res. Board Can. 32: 341–346.

Dans un petit étang de castors peu profond, dépourvu de prédation, près de la baie Georgienne, Ontario, *Ceriodaphnia quadrangula* montre un sommet, et *Daphnia ambigua* et *Bosmina longirostris* trois sommets d'abondance. Chaque espèce atteint un pic et décline rapidement, dépassant probablement la capacité de charge d'un milieu à nourriture limitée. Nous avons déterminé au laboratoire la durée du développement embryonnaire de chaque espèce à différentes températures et avons adapté ces durées à la fonction de Bělahrádek. Les taux instantanés de naissance et de mortalité sont normaux chez *D. ambigua* et *C. quadrangula*, mais trop bas pour expliquer les rapides fluctuations du nombre de *B. longirostris*, ce qui donne à croire qu'il y a eu erreurs manifestes d'échantillonnage. *Bosmina longirostris* peut abandonner périodiquement le limnoplancton et adopter une existence benthique, évitant ainsi d'être capturé.

Received July 8, 1974
Accepted December 3, 1974

Reçu le 8 juillet 1974
Accepté le 3 décembre 1974

MOST studies on natural populations of Cladocera have been carried out on water bodies containing planktivorous predators that strongly influence various dynamic properties of the cladoceran populations, particularly the instantaneous rates of population change and mortality (Hall 1964; Tappa 1965; Wright 1965). In Sunfish Lake, Ontario, where predation was negligible, Clark and Carter (1974) hypothesized that food was the most important single factor controlling population dynamics and, in addition to affecting rates of population change and mortality, also had a strong influence on fecundity (eggs/ovigerous female) and hence on the birth rates of the cladocerans. The present paper examines and contrasts the seasonal cycles and population dynamics of *Bosmina longirostris* (O. F. Müller), *Ceriodaphnia quadrangula* (O. F. Müller), and *Daphnia ambigua* Scourfield in a water body that apparently contains no predators and where cladoceran community dynamics are therefore dependent upon either inherent characteristics of each species or on environmental pressures other than predation.

Printed in Canada (J3484)
Imprimé au Canada (J3484)

Materials and Methods

Hangdog Pond is a small (0.36 ha) shallow (z_m = 1.7 m) beaver pond lying in a Precambrian rock basin on an island in Bayfield Inlet, Georgian Bay, Ont. A thin layer of fine sediments covers the bottom but intense coloration of the water inhibits light penetration and precludes rooted vegetation. The pond is normally ice-free between late April and early December and heats and cools rapidly (Fig. 1). As there is little shelter from prevailing winds the water is continually mixed and temperatures, which are usually 20–25 C June through September, vary little with depth.

Presumably because of its extreme acidity (pH = 4.7–5.1) Hangdog Pond contains neither fish nor predatory cladocerans, insect larvae, amphipods, or rotifers. The cyclopoid copepod *Mesocyclops edax* occurs in quite large numbers but there is no evidence that it ever feeds on Cladocera. In preserved samples

from Hangdog and many similar ponds of the area *M. edax* never has been found ingesting food other than copepodites of its own species or those of the calanoid copepod *Diaptomus reighardi*. There is also no reference in the literature to *M. edax* preying on cladocerans.

Samples were collected on 30 occasions from mid-May through late November, 1974 from four stations in the deepest areas of the pond with a Schindler–Patalas plankton trap of 23 liters capacity equipped with a No. 17 (apertures 0.081 mm) net and collecting bucket. To ensure effective sampling of most of the water column at each station one sample was taken at the surface and a second with the trap about 15 cm above the bottom. The eight samples were then combined, preserved in 95% ethyl alcohol, and later transferred to 10% formalin. Intervals between sampling visits averaged about 5 days during summer and were somewhat longer in spring and autumn. Temperatures were recorded at ½-m intervals at the deepest station on all occasions. In the laboratory each sample was diluted to a volume (500–2000 ml) consonant with the density of zooplankton and 25-ml subsamples were then extracted after thorough mixing. At least 200 (when available) and usually 500 animals of each species were counted, sexed, measured, and examined for eggs and ephippia.

Live animals and pond water were periodically returned to the laboratory in a 22.75-liter plastic container. Nonovigerous females of the three Cladocera were each transferred to a 150-ml pyrex beaker containing 100 ml filtered pond water. Six replicates of each species were then placed in dark incubators at 6, 10, 15, 20, 25, and 30 C. *Chlorella vulgaris* was added to each beaker to a concentration of 0.4 $\times$ 10^6 cells ml^{-1}. Every 6 days at the three colder temperatures and every 3 days at the three warmer temperatures the water was changed and a fresh supply of *Chlorella* added. The animals were observed every 8–12 h at the three colder temperatures and every 4–6 h at the three warmer temperatures; all animals with embryos in the later stages of development were observed more frequently. Development time was considered as the interval between the entrance of the egg into the brood pouch and the moment when the young animal was extruded.

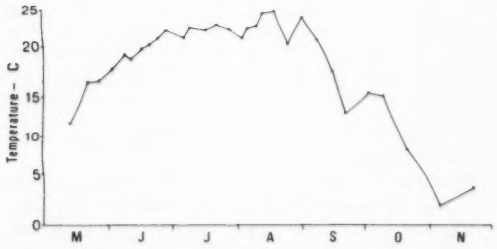


FIG. 1. Mean water temperature in Hangdog Pond May–November, 1973.

Bělahrádek's equation relating development time to temperatures was fitted following McLaren (1963, 1966). This is given by $D = a(T-\alpha)^b$, where D = development time in days, T = temperature C, and a, b, and α are fitted constants.

Results

Embryonic Development

At 10 C, *C. quadrangula* failed to produce any eggs while *B. longirostris* and *D. ambigua* hatched only two and three, respectively; at 6 C no eggs were produced by any species. For the successful experiments the duration of embryonic development and the constants of the equations for the three cladoceran species are given in Table 1 while the computer fitted curves appear in Fig. 2. Each curve was analyzed against each of the other two for variance, slope of the line, and intercept of the line. While variances were not significantly different both slopes and intercepts were highly significant ($P < 0.01$) in every case. Since the curves all had extremely high correlation coefficients (>0.85), indicating low variances, it was considered legitimate to extrapolate them a short distance at each end.

In the high temperature range the three curves lie close together but diverge markedly below 25 C. *Ceriodaphnia quadrangula* embryos develop most rapidly in the 16.5–25 C range but in cooler waters the curve becomes increasingly steep suggesting that the lower temperature limit for development is above 10 C. *Bosmina longirostris* development is somewhat slower than in the other two species over most of the experimental range but at lower temperatures the curve flattens slightly and at 8.5 C crosses that of *D. ambigua* suggesting that *B. longirostris* embryos have the greatest tolerance for coldest waters. Over most of the upper range of temperatures the *D. ambigua* curve falls midway between those of *C. quadrangula* and *B. longirostris*.

Population Dynamics and Seasonal Abundances

In determining instantaneous rates of birth, population change, and death for the pond populations of the three cladoceran species, Hall's (1964) method as applied to *Daphnia galeata mendotae* was employed. The instantaneous birth rate, b, is calculated:

$$b = \ln(1 + B) \quad (1)$$

where B, the finite birth rate is given by:

$$B = N_e/DN \quad (2)$$

TABLE 1. Duration of embryonic development and parameters of development rate $D = a(T - \alpha)^b$ in the three Cladocera.

Species	No.	Mean temp. (C)	Mean duration days	Range	a	b	α
D. ambigua	3	10	8.34	6.93–10.06			
	4	15	4.12	3.97– 4.18			
	9	20	2.47	2.22– 2.75	33.81	–0.976	5.9
	12	25	1.79	1.56– 2.03			
	5	30	1.62	1.42– 1.77			
C. quadrangula	6	15	4.53	4.12– 5.37			
	5	20	2.61	2.35– 2.84	7.178	–0.551	12.4
	7	25	1.76	1.56– 1.93			
	6	30	1.27	1.09– 1.45			
B. longirostris	2	10	10.0	9.50–10.50			
	5	15	5.37	4.91– 7.12			
	6	20	2.99	2.62– 3.25	5631.0	–2.387	–3.9
	6	25	1.60	1.50– 1.79			
	6	30	1.35	1.25– 1.60			

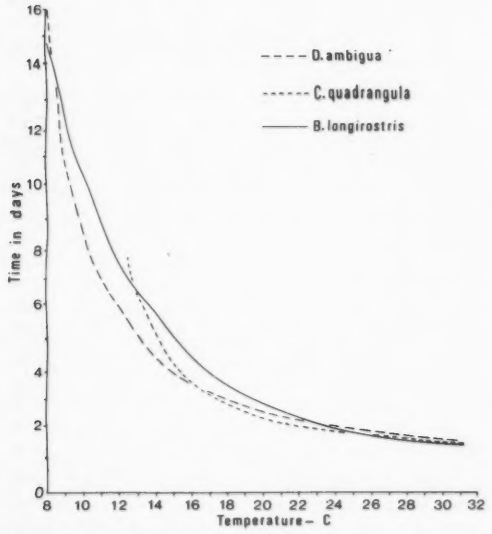


FIG. 2. Duration of embryonic development in *D. ambigua*, *C. quadrangula*, and *B. longirostris* at different temperatures.

in which N_e and N are the number of embryos and the number of animals, respectively, and D is the embryonic development time at the respective temperature. The observed instantaneous rate of population change *r*, is given by:

$$r = \frac{\ln N_t - \ln N_o}{t} \qquad (3)$$

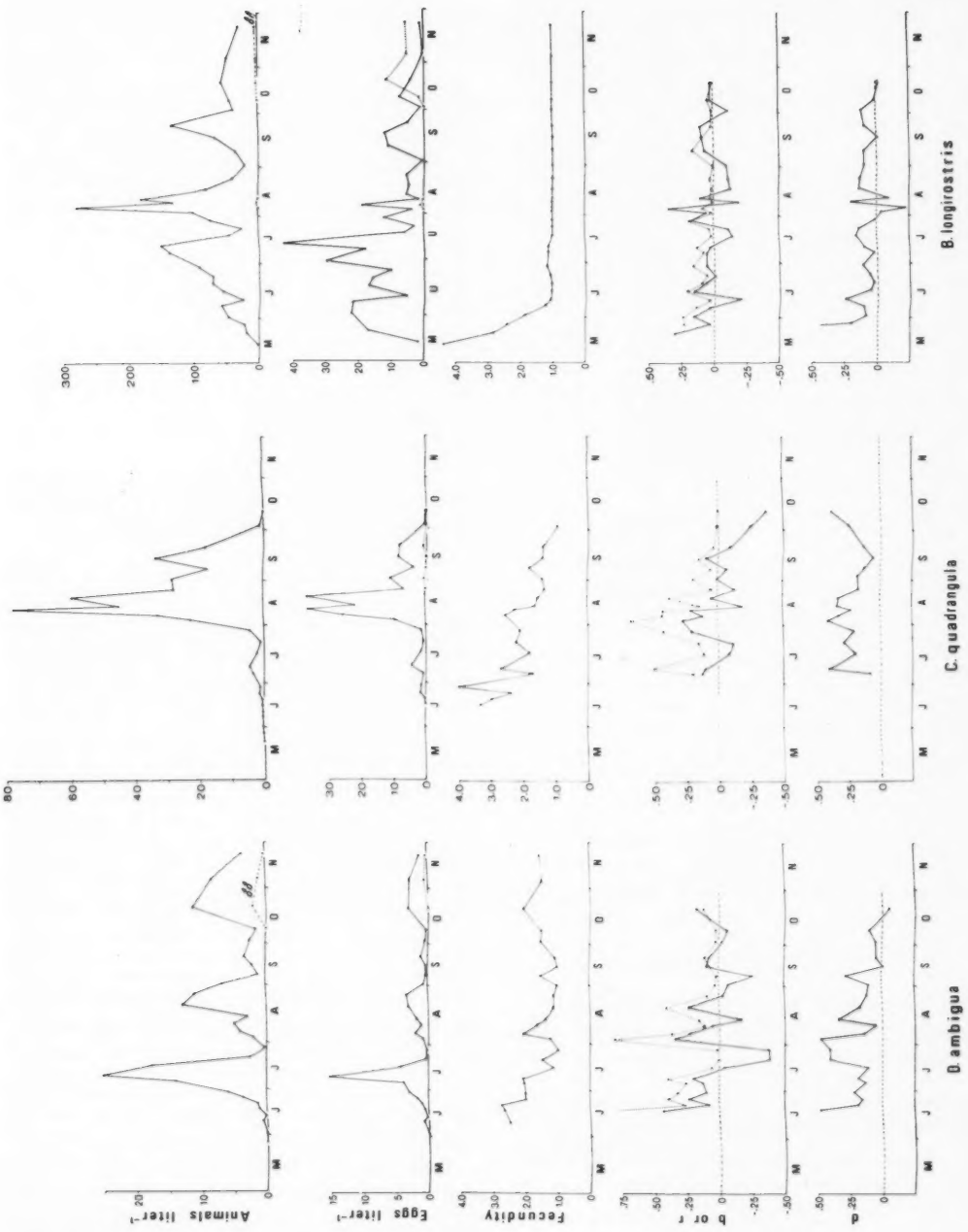
where N_t represents the observed value of N after an interval of *t* days. Knowing *b* and *r* the instantaneous death rate, *d*, may be calculated as:

$$d = b - r \qquad (4)$$

Seasonal abundances of animals and eggs, fecundities and instantaneous rates of birth, population change, and death of the three species are given in Fig. 3. No rates were calculated for dates when a species was scarce in spring or for the two dates in November when in situ temperatures were much beyond the curves of Fig. 2.

Daphnia ambigua appeared first in late May and showed a seasonal cycle of three major pulses. The population expanded rapidly in late June–early July and reached a seasonal maximum of 25 liter^{-1} on July 10. Numbers then immediately fell, the population almost disappearing in late July before reviving in early August and reaching a second peak on August 19. Numbers again declined in late August–early September prior to the final pulse in late October. Males first appeared mid-October. The major egg pulse occurred during the first population peak, subsequent egg production being somewhat lower relative to numbers of animals. A few ephippia appeared only in November. Fecundity of *D. ambigua* oscillated through the season with highest values usually occurring during periods of population increase.

Most intervals of population growth of *D. ambigua* were accompanied by increased birth rates, positive rates of population change and decreased death rates, while population declines were normally reflected in decreased birth rates,


Animals liter⁻¹
Eggs liter⁻¹
Fecundity
b or r
d
B. ambigua
C. quadrangula
B. longirostris
M J J A S O N

negative rates of population change and increased death rates. The two instances of apparent negative death rate in autumn were undoubtedly the result of slight sampling errors at times when asexual egg production was low.

Ceriodaphnia quadrangula was present in the plankton from mid-May, but the population remained low until early August when numbers increased rapidly to a seasonal peak of 77 liter^{-1} on August 10. With some fluctuations numbers then declined until a smaller second peak was attained on September 15. By early October the species had disappeared from the pond. Egg numbers closely paralleled those of the animals with the major peaks in August. A few ephippia appeared in September but males were never seen. Although Lilljeborg (1900) described the male of *C. quadrangula* from Europe there is yet no published record from North America which suggests that the ephippial eggs from the pond may have been pseudosexual. Fecundities were highest during the early season, reaching 4.0 on June 28, after which they declined with minor increases during the August and September pulses.

Instantaneous rates of birth and population change showed marked oscillations with highest values occurring at the time of the August population maximum. The death rate showed little consistent pattern except in autumn when it decreased during the September pulse and then increased with the subsequent population decline; during July and August it was moderately high at all times, values usually exceeding 0.25.

Bosmina longirostris greatly outnumbered the other two cladocerans. Present on the first sampling date, the population increased steadily until mid-June when it declined briefly before reaching the first major peak on July 10. During the latter part of July numbers dropped off sharply prior to ascending to a seasonal maximum of 280 liter^{-1} in early August. After a second decline a final peak was reached in late September. Through October and November numbers first increased slightly then gradually declined to 30 liter^{-1} on November 22. A few males appeared briefly on August 10 and in greater numbers after mid-October.

Bosmina longirostris produced a majority of eggs during the first population increase from May through mid-July. For the remainder of the season production was considerably lower even during the two later population peaks. A few ephippia were seen on August 6 but did not reappear until October 9; on the final three sampling dates ephippia greatly exceeded asexual eggs in numbers. Fecundity was 4.4 on May 13 then decreased rapidly until mid-June. After mid-July fecundity never rose above 1.0 which was largely the cause of the low egg production during the period.

Birth and death rates of *B. longirostris* were generally lower than in the other two species while shifts in rate of population change were less pronounced. Even at times of maximum population increase the birth rate seldom exceeded 0.20 and was usually much less; during population declines it was only slightly above zero. Death rates never exceeded 0.25 even during the steepest population decline. The negative death rates calculated for five dates apparently resulted from extremely low values of b concurrent with high positive values of r.

Discussion

The three cladocerans in Hangdog Pond had basically different seasonal cycles. *Ceriodaphnia quadrangula* was a predominantly summer species, *D. ambigua* was summer and autumnal, while *B. longirostris* occurred in relative abundance throughout the entire period. However, they shared a common tendency for sudden numerical increase followed in each instance by a rapid decline. Presumably this represents an overshooting of the carrying capacity of a food-limited environment lacking the dampening effects of predation. For the most part when numbers of either *D. ambigua* or *C. quadrangula* expanded or declined there occurred the appropriate changes in birth and death rates. *Bosmina longirostris*, on the other hand, though peaking to high numbers on three occasions, showed only minor changes in birth and death rates at any time and even these were not always consistent with the direction of population change. During the 15 days between July 22 and August 6 numbers increased twelvefold yet the birth rate showed only two small upward movements during the period. Mean birth and death rates (Table 2) indicate mean population replacement times in *C. quadrangula* and *D. ambigua* of only about 4 and 5–6 days, respectively. In *B.*

FIG. 3. (*p. 344*) Seasonal abundances of animals, seasonal abundances of eggs (*solid lines*) and ephippia (*dotted lines*), fecundities (mean eggs/ovigerous females), birth rates (*solid lines*) and rates of population change (*dotted lines*), and death rates of *D. ambigua*, *C. quadrangula*, and *B. longirostris*.

TABLE 2. Mean birth rate (b), death rate (d), fecundity (asexual eggs/ovigerous female), and percent females ovigerous (asexual eggs) in each of the three Cladocera.

Species	Mean b	Mean d	Mean fecundity	Mean % ♀♀'s ovigerous
D. ambigua	0.19	0.17	1.67	19.1
C. quadrangula	0.24	0.24	2.11	25.3
B. longirostris	0.11	0.11	1.27	16.7

longirostris replacement time is about 9 days which does not fully accord with its rapid increases and declines throughout the season. That *B. longirostris* probably has a somewhat slower rate of reproduction than the other two species is certainly indicated by its lower mean seasonal fecundity and lower mean percentage of ovigerous females (Table 2); in addition, its embryos require a slightly longer development time at temperatures between 13 and 24 C (Fig. 2), the prevailing summer range in the pond.

There is a strong possibility that the apparent population crashes of *B. longirostris* in July and August were in fact due to gross sampling errors, in which case the validity of the values for *b*, *r*, and *d* (originally derived in part from N) might seem questionable. However, since these values remained constantly low through the declines and subsequent increases in numbers, it is reasonable to suppose that they approximate the actual values for the total population. Continuous sampling errors of such magnitude in a small body of water would have been possible only if the animals had either been aggregating into fairly compact clusters or had temporarily emigrated from the limnoplankton. The former seems unlikely over an extended period though cladoceran aggregation has been noted in Europe by Smyly (1958) and H. B. N. Hynes (personal communication). The more plausible explanation is that the majority of the animals had temporarily abandoned the limnoplankton and, since the pond has no littoral zone, descended to the bottom sediments. Such behavior might be attributed to food shortage in the plankton because *D. ambigua* and *C. quadrangula* both suffered heavy mortality during the same periods. That intense crowding and consequent food depletion occurred during its August pulse is certainly indicated in the production by *B. longirostris* of both males and ephippia (see discussion in Hutchinson 1967). By retreating to the benthos *B. longirostris* might have had access to some form of sedimented detrital food unavailable to *D. ambigua* and *C. quadrangula*; the importance of detritus as zooplankton food has been suggested by many authors (e.g. Edmondson 1957). Seki (1972) has demonstrated that microorganisms, which might have great food value, form an integral part of detritus particles in the sea. It has been noted that planktonic cyclopoid copepods may also become benthic at times (Moore 1939; Comita 1972; Carter 1974). Undoubtedly *B. longirostris* is a versatile animal since it occupies virtually every type of freshwater habitat and is known to migrate between the limnoplankton and littoral as external conditions change (Wesenberg-Lund 1904).

Acknowledgments

This work was supported by a grant from the National Research Council of Canada to J. C. H. Carter. We are indebted to Dr J. C. Roff, University of Guelph for assistance with Bělahrádek's function.

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Tidal and Diurnal Effects on Zooplankton Sample Variability in a Nearshore Marine Environment

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SAMEOTO, D. D. 1975. Tidal and diurnal effects on zooplankton sample variability in a nearshore marine environment. J. Fish. Res. Board Can. 32: 347–366.

Two 26-h series of duplicate and replicate vertical zooplankton samples taken at two stations at two different times of the year showed that a periodic fluctuation correlated to the tide was present in the biomass of zooplankton and in the numbers of animals of several copepod species. Diurnally migrating species of zooplankton did not show a tidal correlation. The hour to hour fluctuations in zooplankton were generally more important than the difference between stations 1.6 km apart. Similar relations were found between the zooplankton and the tide in May and August even though there were significant differences in the composition of the zooplankton community during the two periods.

Correlation coeficients between all species in all samples showed the presence of a diurnally migrating predator community and a nonmigrating prey community consisting of copepods.

An experiment using sample data suggested that to obtain a reasonably accurate estimate of the mean numbers of a given species, one should make at least two vertical tows on a station at a 6-h interval.

SAMEOTO, D. D. 1975. Tidal and diurnal effects on zooplankton sample variability in a nearshore marine environment. J. Fish. Res. Board Can. 32: 347–366.

Deux séries, réparties sur une période de 26 h, de prises verticales de zooplancton effectuées en double ou davantage à deux stations à deux époques différentes de l'année démontrent l'existence de fluctuations périodiques, liées à la marée, dans la biomasse du zooplancton et dans le nombre d'individus de plusieurs espèces de copépodes. Les espèces de zooplancton à migrations diurnes ne montrent pas corrélation avec la marée. Les fluctuations d'heure en heure du zooplancton sont en général plus importantes que la différence entre les stations situées à 1.6 km l'une de l'autre. Nous avons observé des relations semblables entre le zooplancton et la marée en mai et en août, en dépit de différences significatives dans la composition de la communauté zooplanctonique à ces deux périodes.

Les coefficients de corrélation entre toutes les espèces dans tous les échantillons démontrent la présence d'une communauté prédatrice à migrations diurnes et d'une communauté proie non migratrice composée de copépodes.

Une expérience fondée sur les données de l'échantillonnage suggère que pour obtenir un estimé raisonnablement précis du nombre moyen d'individus d'une espèce donnée, on devrait faire au moins deux prises verticales à une station à 6 h d'intervalle.

Received April 29, 1974
Accepted December 9, 1974

Reçu le 29 avril 1974
Accepté le 9 décembre 1974

THE estimation of the abundance of zooplankton in a particular body of water from plankton net samples is subject to many sources of variability. Among these, the most important is variability due to the patchy distribution of virtually all forms of zooplankton (Cassie 1963). This variability creates special difficulties in determining how representative local samples are of the average densities of animals present over a wider area or longer time.

The interpretation of zooplankton variability becomes a more acute problem when a time series of plankton samples is taken at weekly or monthly intervals to measure seasonal trends. The approach is generally to take a small number of replicate vertical or oblique tows on a fixed geographic station. In addition to the variability in samples due to spatial heterogeneity in density and composition, there are diurnal or tidal influences on the zooplankton samples at the stations. The influence and significance of these time factors on the sample variance are little understood, and require special sampling designs for their study.

This report examines the problem of variability

Printed in Canada (J3404)
Imprimé au Canada (J3404)

in numbers of animals of different species in a series of zooplankton samples taken by a vertically towed net over a time period of about two tidal cycles. It compares the pattern of zooplankton variability on two fixed geographic stations during the same tidal cycles and at two different seasons of the year. The occurrence and representation of species in the samples is examined for evidence of correlated "groups."

Methods

Sampling Technique

A ½ m #6 (243 μ) mesh bongo net (McGowan and Brown 1966) with a TSK flow meter fitted in the mouth of each net was used for all tows. The nets were 2 m long from the mouth to the end of the plankton bucket. The bongo net's advantage over the conventional single ring net is that it does not have a bridle preceding the mouth of the net to frighten and warn animals of the net's approach. It takes two samples simultaneously, thereby providing a means of measuring the net sampling error.

There is some question as to whether the two samples taken with the bongo net are independent with respect to space, and if they should be used as a measure of the experimental error. The argument against the net is that because the two nets are attached, they each influence the space sampled by the other. Since one of the objectives of the experiment was to determine the effect of short time intervals on the sample variability, samples used to measure the experimental error had to be made at the same time. Whether samples were collected with separate nets on the same wire, or with nets on different wires from the same boat, one could still argue that the space was not sampled randomly. Therefore, the two sides of the bongo net gave as good an estimate of the sampling error as was possible for the conditions laid down by the experiment.

Vertical tows were made from a depth of 2 m from the bottom to the surface in St. Margaret's Bay, N.S. The depth of the stations maintained during August 11 and 12, 1971, was 35 m and during May 24 and 25, 1972, was 55 m. The net was towed at a speed of 1.5 m s^{-1}.

In each experiment two stations (A and B) 1.6 k apart were marked with floating buoys anchored to the bottom (Fig. 1). A distance of 1.6 k was chosen because Platt et al. (1970) showed a levelling off of the $\log_{10}$ coefficient of variation between phytoplankton samples at about this distance. In addition, this distance did not require an excessive amount of travelling time between stations.

The tows were made on Station A, the second beginning within 4 min of the termination of the first. During August the time needed for a tow was about 1 min, and during May about 2 min. The boat then moved to Station B where two tows were made. The time lapse between the first tow on Station A and the second tow on Station B was 12–15 min. The boat returned to Station A after the tows on Station B. One hour after the first tow on Station A the sampling cycle was repeated. This procedure was repeated hourly for 26 h.

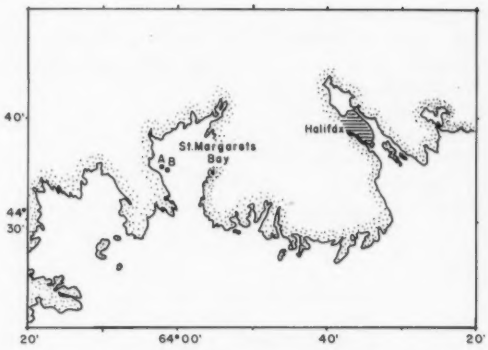


FIG. 1. Sampling stations in St. Margaret's Bay.

Samples were preserved in a 5% formalin and seawater solution. The large zooplankton (chaetognaths, mysids, medusae, ctenophores, and fish larvae) were removed from the samples, identified, counted, and wet weighed. The water was removed from the remaining sample, which consisted mainly of copepods (called zooplankton biomass in the text), by vacuuming over a 30-μ filter until the water ceased to drip. The sample was then weighed to obtain a zooplankton wet biomass estimate. This sample was then split a number of times and one of the portions used for species identification and counts. The reproducibility of the subsampling results were tested by recounting 25 samples. The values differed by an average of 5.1 ± 2.1%. This was a small error compared to the variation between samples and therefore was ignored in the analysis.

The term species component refers to parts of the samples separated for analysis, such as individual species numbers or biomass.

Statistical Analysis

Biomass measurements and numbers of animals in each species were converted to numbers 100 m^{-3} and m^{-3} using the flow meter data. These numbers were then converted to $\log_{10} + 1$ to decorrelate the mean and standard deviation. A hierarchical analysis of variance (Bennett and Franklin 1954) was then calculated for the data. The data were partitioned into four levels as follows:

Level	
1	Time (hours, 1–26)
2	Station (A and B)
3	Tow (1 and 2)
4	Sample (1 and 2)

The time between tows on a particular station (4 min) and the time between stations (12–15 min) was not believed to have had a significant effect on the samples; therefore, the assumption was made for purpose of analysis that all four tows were made simultaneously each hour.

The analysis assumes that the logarithms of the observations were randomly and normally distributed.

The degrees of freedom for the different sample components are as follows:

Source of estimate	df
Between hours	26–1
Between stations	2–1
Hour × stations interaction	(26–1) (2–1)
Between tows (within hours and stations)	(26)2(2–1)
Between samples (within tows, hours, and stations)	26(2(2(2–1)
Total	26(2(2(2)–1

The mean values for the individual zooplankton species component in samples 1 and 2 for each tow were determined and plotted against time for each station. These mean values were used in a Fourier regression analysis with 1–3 harmonics to check for possible periodicity in the data over the 26-h period. This was done separately for each station.

The least squares method was used to fit the data to the following regression equation:

$$Y_t = \alpha_0 + \sum_{i=1}^{n} \alpha_i \operatorname{Cos}(2\pi t/_k)_i + \beta_i \operatorname{Sin}(2\pi t/_k)_i$$

where

α_0 = mean of y

$\alpha_i = \dfrac{\text{Square of products } u_i T_i}{\frac{1}{2}fk}$

$\beta_i = \dfrac{\text{Square of products } v_i T_i}{\frac{1}{2}fk}$

$u_i = \operatorname{Cos}\left(\dfrac{2\pi}{k}t\right)_i$

$v_i = \operatorname{Sin}\left(\dfrac{2\pi}{k}t\right)_i$

f = number of replicates (in this case two)

T_i = total of y at each t

k = number of time units per cycle (a variable depending on the number of cycles being fitted to the data)

t = time units (h)

For a complete explanation of the analysis see Bliss (1958) and Dixon (1968).

The generalized analysis of variance table for the two-term Fourier analysis is as follows (modified from Bliss 1958):

Row	Term	df	Sum of Squares, (SS = S_t)
1	Between replicates	2−1	$\Sigma T_{r2}{}^2/k - C_m$
2	Effect of (a_1+b_1)	2	$\{[u_1T_t]^2+[v_1T_t]^2\}/\frac{1}{2}fk = S_2$
2	Effect of (a_2+b_2)	2	$\{[u_2T_t]^2+[v_2T_t]^2\}/\frac{1}{2}fk = S_3$
4	Scatter about curve	26−5	$\Sigma T_t{}^2/f - C_m - S_2 - S_3$
5	Replicate × (a_1+b_1)	2(2−1)	$\Sigma\{[u_1y]^2+[v_1y]^2\}/\frac{1}{2}k - S_2 = S_3$
6	Replicate × (a_2+b_2)	2(2−1)	$\Sigma\{[u_2y]^2+[v_2y]^2\}/\frac{1}{2}k - S_3$
7	Replicate × scatter	(2−1)(26−5)	By difference
8	Total	(2)26−1	$\Sigma y^2 - C_m = [y^2]$
9	Correction for mean	1	$\Sigma^2 y/fk = C_m$
10	Replicate × A_1	2−1	$2S_2+S_5-2\sqrt{S_2}\,\Sigma\sqrt{[u_ty]^2+[v_ty]^2}/\sqrt{\frac{1}{2}fk}$
11	Replicate × Phase_1	2−1	$S_5 - S_{10}$

This analysis was calculated on data transformed to $\log_{10}(x+1)$ and data not transformed. The curves given in the results are nontransformed because the transformation did not change the significance of any of the terms.

Correlation coefficients (r) were calculated for all possible pairs of sample components taken on each of the stations to determine if any significant r values existed between the different species over the 26 h of the experiment. The data used were the means of the four samples taken on a station each hour. These data were first transformed to $\log_{10}(x+1)$. One was added to all the values before the $\log_{10}$ transformed to eliminate 0 values.

Results and Discussion

August Experiment

A series of paired t tests between samples 1 and 2 showed no significant difference between any of the zooplankton components in samples taken on the same tow. Therefore, there was no

TABLE 1. Degrees of freedom and values of F for various zooplankton components in the hierarchical analysis of variance for August 1971 experiment.

Sources of estimate	df	No. *Sagitta elegans*	Wt *Sagitta elegans*	No. *Sagitta* eggs	No. Cunner	No. *Podon*	No. *Evadne*	Wt Zooplankton	No. *Oithona*	No. *Eurytemora*	No. *Calanus*	No. *Acarita*	No. *Temora*	No. *Pseudocalanus*	No. *Mysis mixta*	Wt *Mysis mixta*
Between hours	25	8.38*	6.84*	0.98	2.30*	2.92*	8.43*	3.99*	8.44*	7.24*	3.04*	6.93*	7.44*	5.39*	18.16*	12.07*
Between stations	1	1.97	6.06*	9.18*	1.53	0.77	23.76*	0.88	2.07	1.36	9.39*	2.01	8.60*	2.16	0.62	0.52
Hours × stations	25	3.05*	2.68*	3.49*	1.21	0.95	1.12	2.81*	0.81	1.09	0.90	0.65	2.52*	1.82*	1.10	1.25
Between tows within stations	52	1.56*	1.80*	0.34	1.05	1.25	1.03	1.22	0.81	0.92	0.83	0.78	0.60	0.53	7.37*	9.36*
Between samples within tows	104															

*F values significant at .05 level.

difference in the catching efficiency of the two sides of the net.

The hierarchical analysis of variance showed no significant difference between successive (duplicate) tows for any sample component except *Mysis mixta* biomass and numbers and *Sagitta elegans* numbers and biomass (Table 1).

Six species components had significant interactions between hours and stations, which meant the changes with hours were influenced by the station on which they were measured.

The biomass of *S. elegans*, numbers of *S. elegans* eggs, and numbers of the cladoceran *Evadne* and copepods *Calanus* and *Temora* differed significantly ($P < .05$) between stations. All components of the samples were significantly different between hours except the number of *S. elegans* eggs (Table 1).

All components of the samples with a significant difference between hours on Station A except the larvae of cunner (*Tautogolabrus adspersus* Walbaum with a mean length of 7 mm) showed a significant first or second harmonic regression (Fig. 2), and of these only the biomass of *S. elegans* and the numbers of *Evadne* did not show a significant reduction in the mean square for the scatter about the curve when the second harmonic was removed from the data. The copepod *Oithona* was the only component with a significant third harmonic periodic regression.

A similar statistically significant ($P < .05$) periodic regression pattern was found for the components of the samples taken on Station B. The main differences were in data for the copepod species *Oithona* which did not have a significant second harmonic regression, but had a significant third harmonic regression, and the presence of a significant interaction between replicates and the first harmonic for the numbers of the copepod *Calanus*.

These results meant that fluctuations in biomass and numbers with time, although not identical for each species, were nonrandom for all but cunner larvae on Station A. The same harmonic pattern was found for data of both replicate series for all sample components except *Calanus* on Station B, which indicated that the data for *Calanus* were more variable on Station B than on Station A.

The first, second, and third harmonics accounted for 10–85% of the total variance in the Fourier analysis (Table 2), depending on the species component of the sample. The least reduction in the total variance occurred in the data for the cunner larvae on Station A and the maximum reduction for numbers of *S. elegans*.

The species composition in St. Margaret's Bay was different in May than in August (Tables 1 and 3). Large medusae and ctenophores were abundant and *S. elegans* individuals were much larger (up to 30 mm in length). The cladoceran, *Podon*, was absent and *Evadne* was present in low numbers. Large numbers of a small species of medusa were present in May that were not found in August.

The numbers of copepods were much lower in May than in August, with the dominant species being *Pseudocalanus* and *Calanus*. The other species that were found in August were present at very low densities.

The two most common species of fish larvae in the Bay were the Atlantic herring (*Clupea harengus harengus*) with a mean length of 30 mm and the sand lance (*Ammodytes* sp.) with a mean length of 17 mm.

A series of paired *t*-tests calculated for each component in the pairs of samples from each tow showed no significant difference between any of the components, that is, the two sides of the net were again equally efficient in capturing all sample components. The hierarchical analysis of variance was then calculated for each of the sample components.

The biomass of *S. elegans* and number of *S. elegans* eggs were the only parts of the sample

TABLE 2. Percentage of total variance removed by first, second, and third harmonic regression.

Zooplankton component	August		May	
	Station			
	A	B	A	B
Zooplankton biomass	66[b]	56[b]	46[b]	43[b]
Pseudocalanus	44[c]	48[c]	33[b]	54[c]
Calanus	34[b]	34[b]	42[b]	17[c]
Oithona	48[c]	56[c]	26[b]	27[b]
Acartia	33[a]	32[b]	31[b]	31[b]
Temora	63[b]	55[b]	70[b]	60[b]
Eurytemora	69[b]			
Evadne	43[a]	63[b]	46[a]	56[b]
Podon	38[b]	32[b]		
Sagitta no.	85[b]	66[a]	79[a]	79[b]
Sagitta biomass	81[a]	59[a]	80[a]	81[b]
Sagitta eggs			47[a]	53[a]
Small medusae			16[b]	22[b]
Cunner larvae	20[b]	10[b]		
Sand lance larvae			21[b]	55[a]

[a]Total variance removed by one harmonic.
[b]Total variance removed by two harmonics.
[c]Total variance removed by three harmonics.

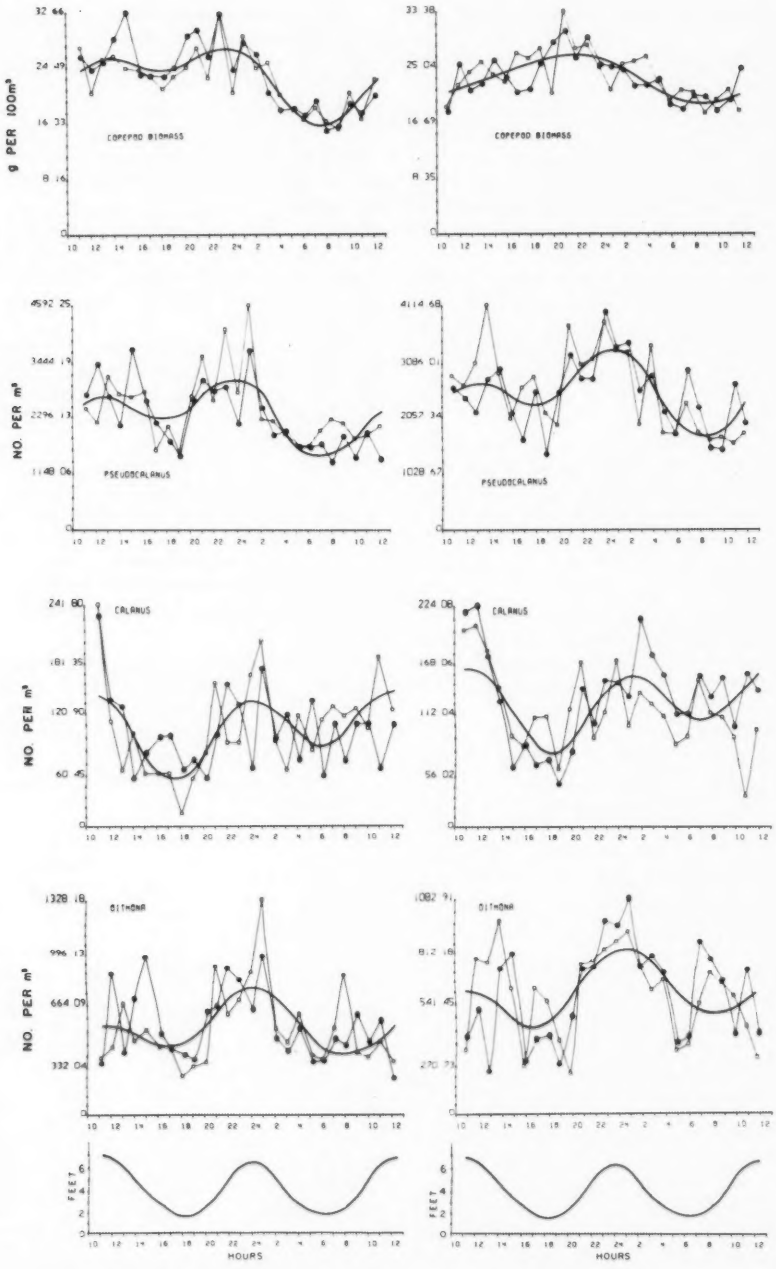
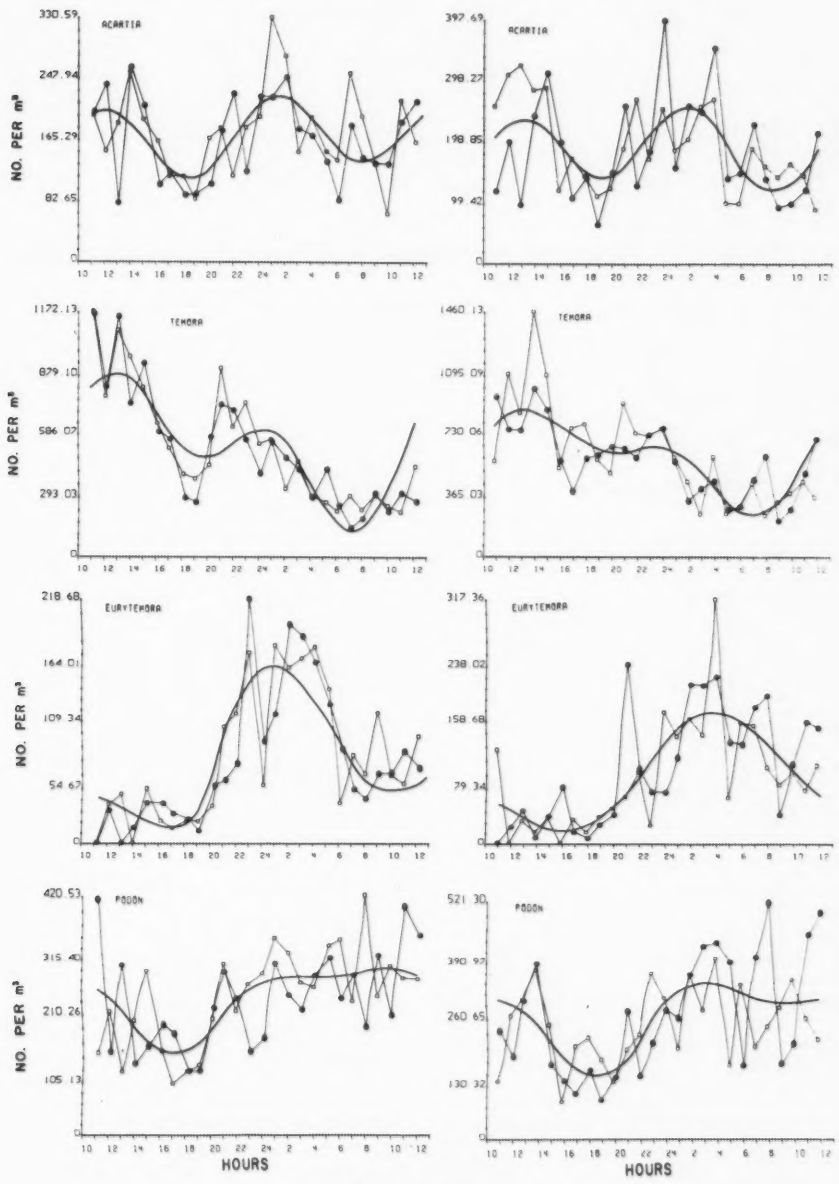
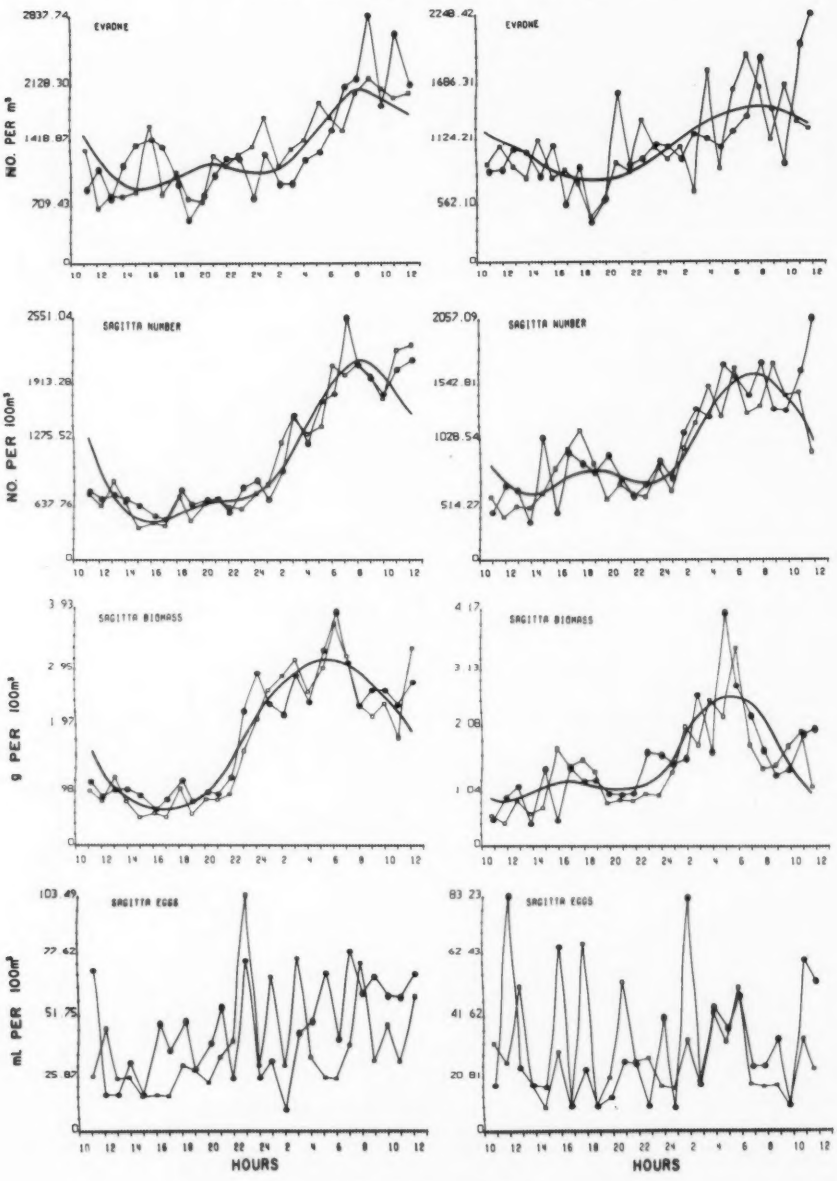


FIG. 2. Mean values of August sample components for tow 1 (○) and tow 2 (●) and fitted periodic regression curves and tidal fluctuation curve for 26 h. Left side is Station A data and right side Station B data. (*Continued*)



(Fig. 2. Continued)

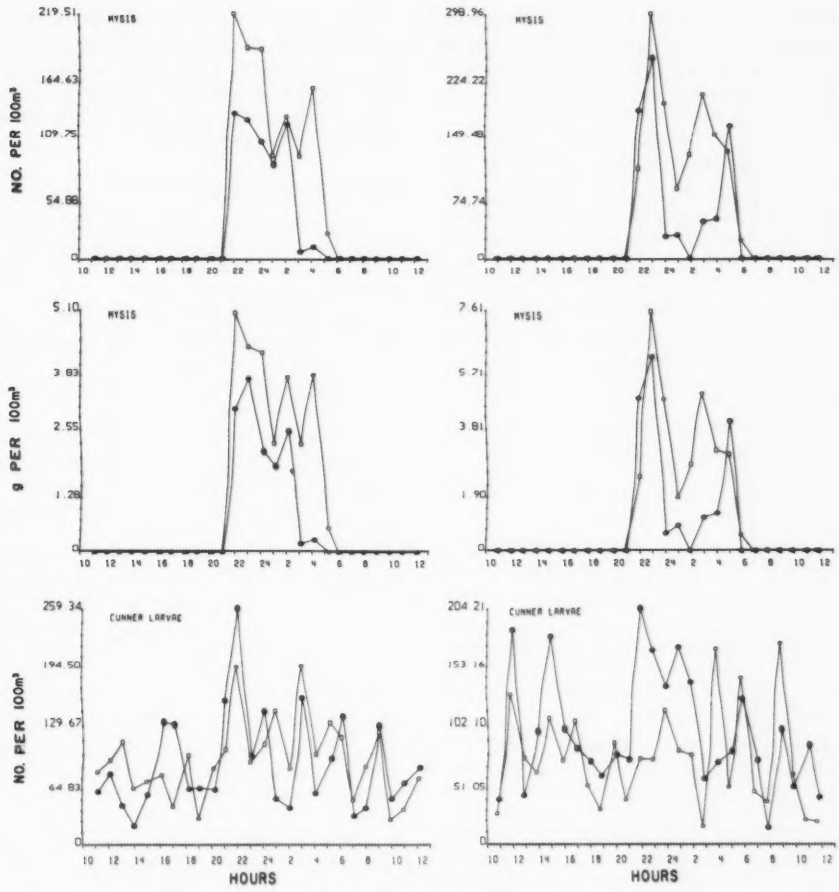
that showed a significant variation between the replicated tows. Five species components, the numbers and biomass of *S. elegans*, milliliters of medusae, and numbers of *Pseudocalanus* and *Calanus* had significant interactions between hours and stations. Three components, *S. elegans* number and biomass and number of *Oithona* had significant differences between stations, indicating there was a difference in the size of the populations on the two stations. Seven of the eighteen components of the samples did not show a significant difference between hours; these were the



(*Fig. 2. Continued*)

volume and numbers of large medusae (>1 cm diam), and the numbers of *Evadne*, *Oithona*, *Acartia*, young *S. elegans*, and herring larvae. This showed there was no significant trend between the numbers of these animals and time.

The Fourier analysis showed all the components of the samples taken on Station A, except numbers of young *S. elegans*, small medusae, large medusae, *Acartia*, and *Oithona*, had a significant periodic regression with time. The numbers of *S. elegans*, *S. elegans* eggs, and *Pseudocalanus* had a significant interaction between the replicates.



(*Fig. 2. Concluded*)

But both replicates showed the same periodic pattern.

The components of samples from Station B all had a significant first or second harmonic regression with time, except the numbers of *S. elegans* eggs, *S. elegans* young, and the copepods *Oithona* and *Calanus*. The data for the biomass of copepods and for most of the copepod species from each station had a significant scatter about the curve after the second harmonic was removed. This scatter was reduced to nonsignificance in *Pseudocalanus* and *Calanus* when the third harmonic was removed. No other components in samples from either station showed a significant harmonic higher than the second. This indicated a more complex pattern of periodic fluctuation for these two copepod species than for the other species components. The percentages of the total variance removed by the harmonics for the May data are given in Table 3. There was little difference in the amount of variance reduction in May and August data.

The harmonic regression removed up to 85% of the variation with time in samples of *S. elegans* and up to 66% of the variance in the data for copepod numbers. The remaining variance was due to variations in animal density too small to be sampled on a time scale of one hour.

Sagitta elegans eggs was the only component that failed to show significant differences between hours in August. It showed irregular variations about the mean with time, and no evidence of cyclical pattern in numbers. This suggested a rather widely dispersed distribution, which is effectively random on the sampling time scale used. The contrast between results for these "pas-

sive" particles relative to the swimming zooplankton is, however, striking. It tempts one to speculate that factors contributing to patchiness include activity of the organisms different from that of the turbulence field in the surface waters.

Sagitta elegans eggs in May showed a significant periodic regression that appeared related to the numbers of *S. elegans* adults. The increased number of eggs with the increase in the number of *S. elegans* may have been due to the animals' spawning, May being one of the main spawning months for the species (Sameoto 1971*a*). But even with this relationship between eggs and adults, the numbers of eggs between samples was more variable than the numbers of animals as shown by the reduction of the total variance by the harmonic regressions for the two components. This suggests that the environment tended to randomize passive components of zooplankton.

Tidal Fluctuations and Zooplankton Periodicity

To determine if a statistically significant relationship existed between the species components and the tide, correlation coefficients (r) were calculated between the tidal heights and data for each species component each hour. The tidal heights were then advanced 1 h and the r values recalculated. This was repeated until the tide had been advanced 4 h.

The largest number of significant r values occurred in the August data when the tide was advanced 1 h. No significant correlation occurred beyond a tidal shift of 2 h (Table 4). The zooplankton biomass and *S. elegans* numbers showed significant negative correlations with the tide on Station B, but no correlation on Station A. Even though few of the components had significant r values with the tide, the calculated periodic regression lines for most of the copepod species appeared to follow the same pattern as the tide with time.

The numbers of the cladoceran *Podon*, followed the tidal cycle for the first 12 h, then appeared to behave independently of the tide. *Sagitta elegans* numbers and biomass did not follow the tidal cycle (Fig. 2).

The May data had more components with significant r values with the tide or one of the tidal height shifts than did the August data (Table 4). Copepod species were the main components having positive correlations with the tide. The largest number of significant r values occurred with a 0 shift in the tidal height. As the tide values were shifted forward fewer components were correlated with the tide (Table 4).

These results showed that the movement of concentrations of animals past the sampling stations was related to the tidal movements of the water. It was not known if the same patch of zooplankton moved past the stations on both the incoming and outgoing tide.

Diel Effect on Zooplankton

In the August samples only *Mysis mixta*, *S. elegans*, the copepod *Eurytemora*, and the cunner larvae showed an increase in numbers during the night and a decrease at dawn.

Mysis showed the most marked increase in density during the night. There appeared to be a drop in density at about 2400 and an increase about 0400 and then a decline to 0 animals per sample at 0600 (Fig. 2).

The drop in numbers of *Mysis* caught as dawn approached can be explained by the animals migrating to the bottom and the increased chance of net avoidance as more light penetrated the water.

During May, *S. elegans* biomass, numbers, and eggs and the larvae of sand lance showed an increase in numbers 100 m^{-3} during the night with the peaks occurring about 0100 (Fig. 3).

The existence of a diurnal pattern in some of the sample components such as sand lance larvae, *S. elegans*, and the copepod *Eurytemora* was surprising, since the net sampled all the depths of water except the last 2 m. This meant that these organisms were either within the bottom 2 m of water during the day and therefore below the mouth of the net or else they were able to avoid the net. It is unlikely that *Eurytemora* could swim fast enough to avoid the mouth of the net. These data support the suggestion made by Sameoto (1971b) that sampling for macrozooplankton and fish larvae be done at night and in addition, it shows that night sampling may also be necessary for an accurate estimate of numbers of copepods. The results showed that for diurnally migrating animals, samples should be taken at the same time each night, preferably between 2400 and 0300 when the peak in numbers and biomass of these animals occurred.

Correlation Between Zooplankton Components

The significant correlations between species components were divided into two groups, those that were significant on both stations and those that were significant on only one station. Due to a 5% chance of a significant correlation on a single station, only species that were significantly

TABLE 3. Degrees of freedom and values of F for various zoopl

Sources of estimate	df	No. *Sagitta elegans*	Wt *Sagitta elegans*	ml medusae	No. medusae	No. ctenophores	ml ctenophores	No. you *Sagitt*
Between hours	25	9.40*	5.18*	1.13	1.00	2.33*	2.64*	1.61
Between stations	1	26.31*	22.42*	0.46	0.00	0.37	0.33	0.13
Hours × stations	25	2.21*	3.01*	2.08*	1.49	0.63	0.58	1.50
Between tows between stations	52	1.09	1.59*	0.60	0.82	1.19	1.07	0.94
Between sides within tows	104							

*F significant at .05 level.

plankton components in the hierarchical analysis of variance for the May 1972 experiment.

oung *tta*	No. *Sagitta* eggs	No. *Evadne*	No. *Temora*	No. *Pseudocalanus*	No. *Calanus*	No. herring larvae	No. sand lance larvae	No. young medusae	Wt copepods	No. *Acartia*	No. *Oithona*
1	7.52*	1.27	2.50*	6.29*	3.42*	1.29	8.02*	4.05*	6.05*	1.70	1.88
3	1.76	0.12	0.17	1.55	0.06	0.02	1.06	1.01	1.85	0.00	5.99*
0	0.55	1.04	1.02	1.78*	1.80*	0.93	0.55	1.42	1.57	1.50	0.79
4	1.86*	1.12	0.91	1.07	0.95	1.23	0.98	0.75	0.85	0.85	0.99

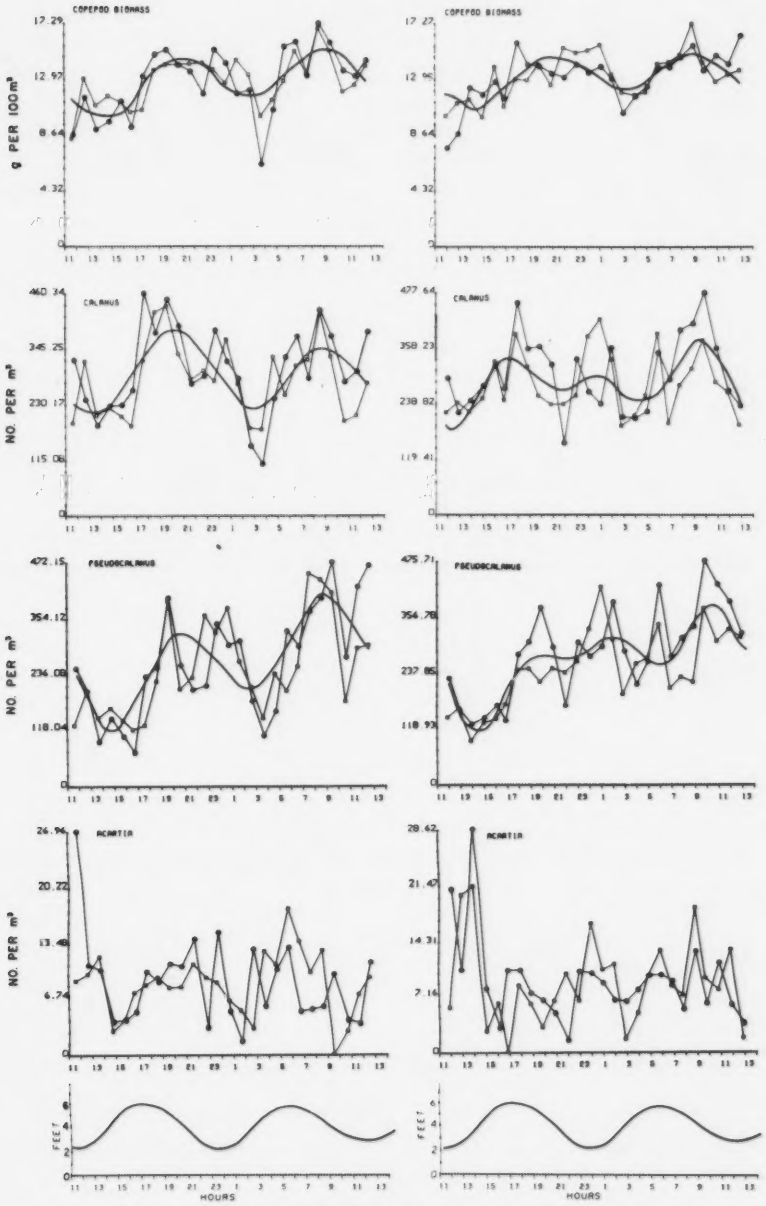
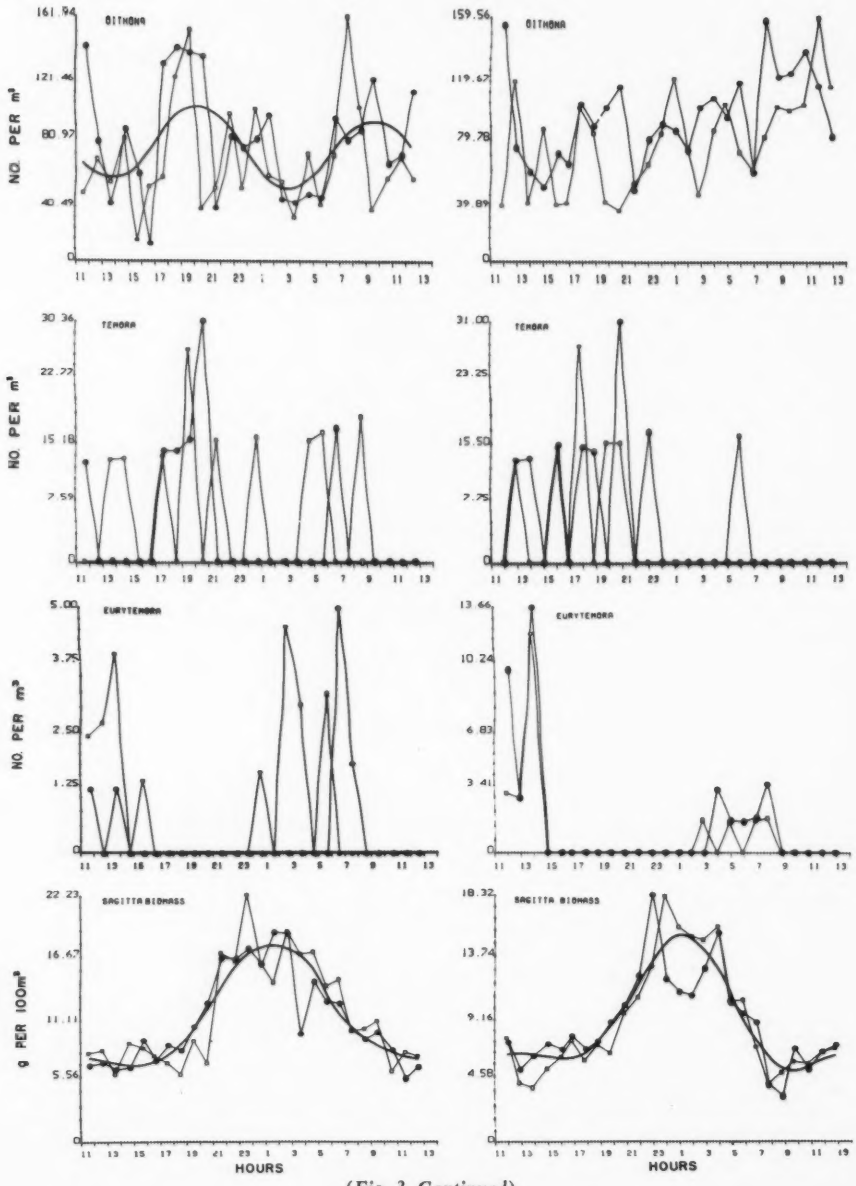


FIG. 3. May data. For explanation see Fig. 2. (*Continued*)

correlated on both stations were used to describe species groups. The probability that the same two species would be correlated by chance on two stations was 0.25%. The pairs of components that were significant on both stations in August had two basic zooplankton component groupings. The first appeared to center around the zooplankton biomass and copepod species, all of

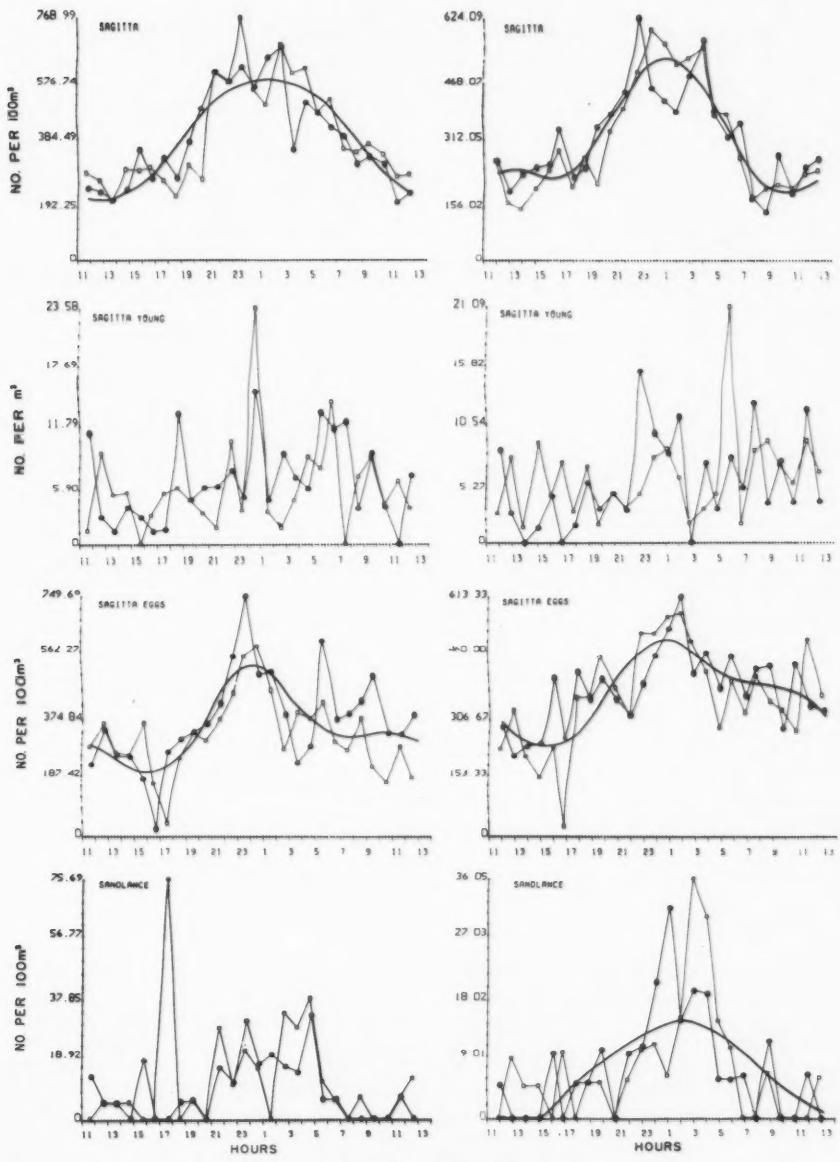


(Fig. 3. Continued)

which were positively correlated with each other, with the exception of *Eurytemora*, which was negatively correlated with *Temora* (Fig. 4). The *S. elegans* biomass was negatively correlated with the zooplankton biomass.

The second grouping in August was centered around *S. elegans* numbers. The copepod species *Pseudocalanus*, *Temora*, and the zooplankton biomass were negatively correlated with *S. elegans* numbers, while the cladocerans *Podon* and *Evadne* and the copepod *Eurytemora* were all positively correlated to either the *S. elegans* numbers or biomass.

Thirteen of the 15 zooplankton components in

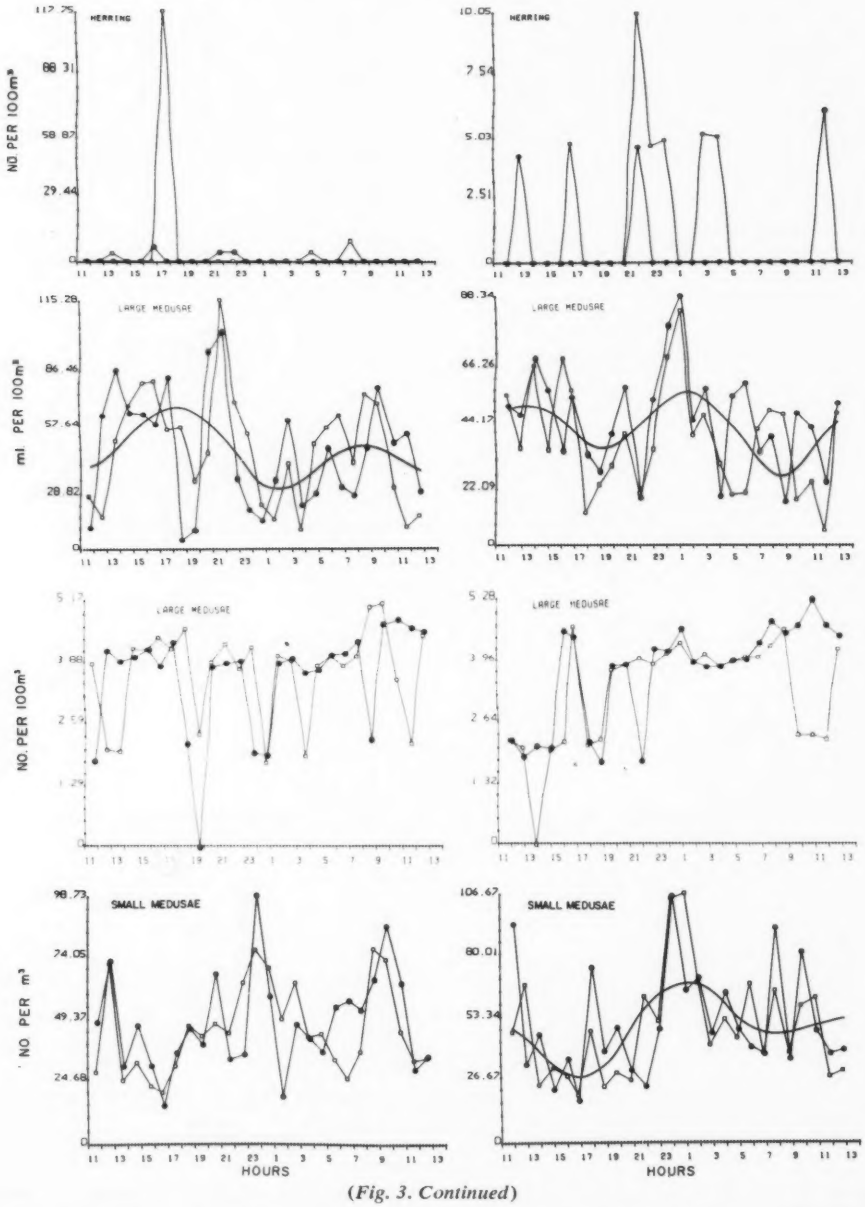


(Fig. 3. Continued)

the August samples had a significant correlation with one or more of the other components on both stations.

The May samples had fewer significant r values between components than the August samples. Ten out of 19 components had significant r values on both stations (Fig. 5). *Pseudocalanus* and *S. elegans* eggs had five significant r values each with other sample components and appeared to be the centers of relations between components. There were no significant negative values of r.

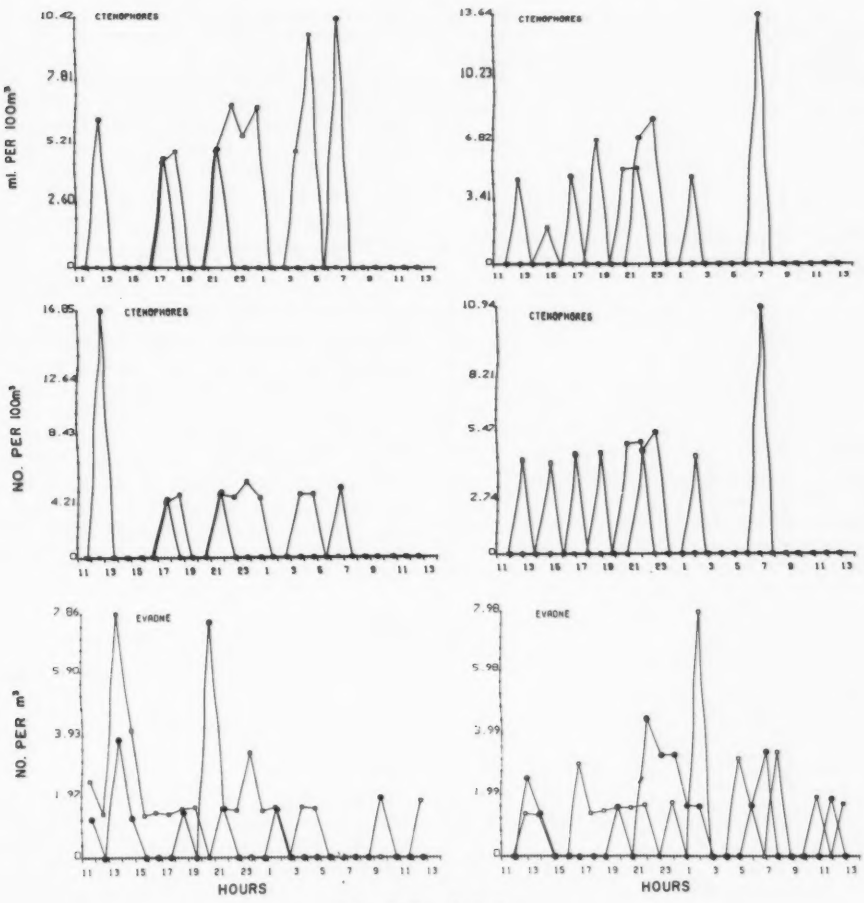
The most striking feature of both the August and May data was the low number of significant correlations common to both stations. Out of a possible 420 combinations of r between the August components only 24 had significant cor-



(Fig. 3. Continued)

relations on both stations and for the May data only 14 out of 648 *r* values were significant on both stations.

Cassie (1963) reviewed the literature dealing with correlation coefficients between zooplankton species and concluded that no consistent pattern in the coefficients could be ascertained, for they could change in magnitude or sign from day to day. He stated that when correlations did exist it was because the animals were reacting to one another or to environmental factors that they responded to in common.



(Fig. 3. Concluded)

The August data had two basic species groupings; the first was made up of the copepods positively correlated with each other and centered around *Pseudocalanus*. This could be considered the prey community. The second grouping, the predator community, centered around *S. elegans* and consisted mainly of copepod species negatively correlated with *S. elegans* numbers or biomass. This negative correlation may have been due to a predator–prey relationship between the copepods and *S. elegans*, or may have resulted from the chance occurrence of the diurnal cycle of *S. elegans* and the tide related cycle of the copepods being out of phase. The copepod *Eurytemora* was positively correlated with *S. elegans*. This was likely due to the increase in their numbers during the night and probably had no biological relationship with *S. elegans*.

Only two copepod species were correlated with the zooplankton biomass, *Temora* and *Pseudocalanus*. This fact and the fact that they were the two most numerous species present suggested they were the main species contributing to the biomass.

The correlation relationships between sample components were simpler in May than in August. There were no significant negative correlations in May. The zooplankton biomass was correlated with only two species of copepods, *Calanus* and *Pseudocalanus*, the two most important species making up the biomass.

It was expected that the fish larvae would have been correlated with the copepods, their main source of food. But this was not so. The herring larvae were not correlated with any of the components, this likely being due to the low numbers of larvae caught. The sand lance larvae correlated

TABLE 4. Zooplankton components showing significant correlation coefficient r ($P < .05$) with the tidal height as it was shifted from 0 to 4 h ahead of the actual times. Significant value of r at 24 df and $P < .05$ is .38.

August			
		r	
		Station	
Tide shift hours	Zooplankton component	A	B
0	*Acartia*	.51	–
	Calanus	.66	.52
	Oithona	.38	–
	Zooplankton	–	–.43
1	*Acartia*	.58	.54
	Temora	.53	.40
	Calanus	.50	.51
	Evadne	.39	–
	Pseudocalanus	–	.43
	Sagitta no.	–	–.39
2	*Acartia*	.54	.59
	Calanus	–	.39
0	Zooplankton biomass	.60	.61
	Pseudocalanus	.45	.39
	Calanus	.67	.40
	Oithona	.39	–
	Sand lance larvae	.39	–.44
	ml medusae	–	–.44
	No. large medusae	–	–.49
1	Zooplankton biomass	.70	.70
	Pseudocalanus	.66	.52
	Calanus	.71	.40
	Oithona	.51	–
	Sand lance larvae	.39	–.43
2	Zooplankton biomass	.69	.60
	Pseudocalanus	.74	.57
	Calanus	.57	–
	Small medusae	.47	–
	Oithona	.49	–
	Acartia	.38	–
3	Zooplankton biomass	.43	.39
	Pseudocalanus	.59	.49
	Acartia	.49	–
	Small medusae	.41	–
	Sand lance larvae	.43	–
	Sagitta eggs	–	.41

with *S. elegans* numbers and biomass, this being the result of the diurnal migration shown by the two species and not because of a predator–prey relationship.

NUMBER OF SAMPLES NEEDED TO ESTIMATE THE POPULATION MEAN

It is not economically or logistically feasible to sample as frequently during a routine sampling program as in this experiment. Therefore, to determine the lowest number of samples during a tidal cycle needed to get an acceptably accurate estimate of the population mean, the following experiment was conducted on the data for species showing a periodic fluctuation related to the tide.

A sample of data of species components that did not show evidence of vertical migration was chosen at random from the 26-h series of data. Beginning with this sample, succeeding samples were drawn from the data series at fixed intervals of 2, 3, 4, or 6 h. Each group of samples started at a new randomly determined time so that the total time from the first to the last sample was 12 h. This ensured that a high and low tide period would be included in the sampling period. In addition a series was run where one sample was taken at random from the 24-h data series. An example of one of each of the five sampling regimes follows:

Time interval (*h*)	Hour when samples taken					
2	0100	0300	0500	0900	1100	1300
3	0300	0600	0900	1200		
4	1100	1500	1900			
6	0700	1300				
12	0900					

The sampling intervals were all less than the time of one zooplankton cycle, thereby preventing aliasing (Platt and Denman 1975). The means of the data selected for each of the time intervals were calculated and the percent deviation (PD) from the mean of 26-h series for that species component calculated.

$$PD = \left(\frac{\bar{X} - \bar{\bar{X}}}{\bar{\bar{X}}}\right) \times 100$$

$\bar{X}$ = mean of sample chosen from the 26-h data series

$\bar{\bar{X}}$ = mean of total 26-h data series

Tests were run separately for each species component on each of the stations. The mean PD (Table 5) for the species components for each of the sampling regimes were compared by a Duncan's multiple range test. There was no significant difference ($P < .05$) in the PD for the numbers of copepods or zooplankton biomass for any of the sampling regimes except that of

TABLE 5. Results of sampling experiment using data collected over 26 h.

Number of samples taken in 12 h	Time interval between samples	% deviation from mean value calculated from data taken hourly for 26 h	
		Copepod numbers	Zooplankton biomass
6	2	17.4±10.2	8.9± 5.9
4	3	13.7± 6.4	7.7± 3.9
3	4	18.6± 6.3	10.9± 5.1
2	6	19.4± 6.1	7.4± 3.5
1	24	41.2±36.0*	17.0±14.1*

*Significantly different from other values as tested by the Duncan's Multiple Range Test.

FIG. 4. Zooplankton components of August data with significant ($P < 0.05$) correlation coefficients on both stations A and B.

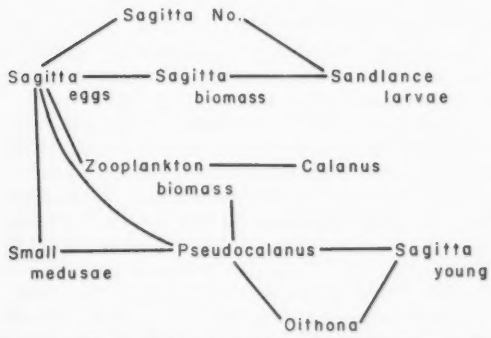


FIG. 5. Zooplankton components of May data with significant ($P < 0.05$) correlation coefficients on both stations A and B.

one sample taken in 24 h. Samples taken at this frequency had a significantly higher PD than any of the other sampling intervals. This demonstrated that it was possible to significantly increase the accuracy of the estimate of the mean zooplankton biomass and the mean for the numbers of different copepod species by taking two samples 6 h apart rather than sampling a station once in 24 h. But sampling more frequently, up to once every 2 h for 12 h, did not make an appreciable difference to the accuracy of the estimate of the mean. This sampling strategy did not apply to organisms that migrate to the bottom during the day or to organisms that demonstrated increased net avoidance during the daytime such as *Mysis*. To sample these species one would have to confine the sampling to the hours of darkness.

VARIATION IN ZOOPLANKTON COMPONENTS

On both stations the arithmetic mean and $\log_{10}$ standard deviation were calculated for each of the 52 sets of four samples taken each hour. The means of the 52 means and $\log_{10}$ standard deviations were then calculated for each of the species components.

The 95% confidence limits for a single observation in the series for each species component was calculated by multiplying and dividing the mean by the $\log_{10}$ standard deviation and calculating the percent of the mean these values represented (Table 6). The confidence limits for copepods were similar in August and May. The limits were considerably narrower than those given by Wiebe and Holland (1968) and Wiebe et al. (1973) for copepods. This may have been due to plankton nets being used with bridles and towing lines preceding the nets, thereby allowing for greater avoidance by the copepods. It is interesting that the passive components of the samples in this study, *Sagitta* eggs, had almost

TABLE 6. Confidence limits of a single observation for various species in August and May samples.

Species	95% confidence limits of a single observation represented as percentage of the mean	
	August (%)	May (%)
Zooplankton biomass	91–110	88–113
Pseudocalanus	76–132	75–134
Calanus	58–173	78–129
Oithona	69–145	69–146
Acartia	63–158	46–213
Temora	77–138	72–138
Evadne	30–333	32–182
Sagitta numbers	31–322	83–115
Sagitta biomass	95–104	83–120
Sagitta young		45–226
Sagitta eggs	30–336	60–165
Podon	60–151	
Cunner larvae	45–215	
Mysis number	20–470*	
Mysis biomass	56–177*	
Small medusae		65–154
Large medusae vol.		63–158
Large medusae no.		30–347
Sand lance larvae		29–341
Herring larvae		53–189

*Night samples only.

identical confidence limits with those of *Sagitta* eggs reported by Wiebe et al. (1973), which were 28.9–346%. This suggests greater avoidance by the copepods in studies reported by Wiebe than in the present study.

When calculating the confidence limits for *Mysis*, only samples taken between 2100 and 0700 were used, because during other times no *Mysis* were caught. Therefore, the values given are confidence limits mainly for night samples.

An interesting observation in August was that the $\log_{10}$ standard deviation for *Mysis* numbers and biomass was significantly higher for tow 2 than for tow 1. This meant that the samples taken on the second tow were more variable than those taken on the first tow. This may have been the result of the animals becoming more wary after being disturbed during the first tow.

General Discussion

The most striking feature of both the August and May data was the evidence of periodicity and the relationship between their fluctuations and the tide. Platt and Conover (1971) found a similar periodicity in the chlorophyll concentration and zooplankton biomass in the mouth of a small marine basin. They found the fluctuations were out of phase with the tide by about 2.5 h. A second experiment in the center of the basin showed a similar fluctuation related to the tide, but this time with a lag of about 4–5 h. Their second experiment lasted only 12 h, from 1200 to 2400; therefore, it was not possible to separate the effect of the vertical migration of the zooplankton from the tidal effect.

In the present study, the samples with the highest and the lowest zooplankton biomass on a station differed by a factor of 2 in August and 2.7 in May. These values are similar to the mean difference factor of 2.1 found by Sameoto (1971b) for zooplankton biomass values taken on five different stations in Bedford Basin, a small marine basin near St. Margaret's Bay. The mean difference factor in the present study for copepod species showing a periodic fluctuation was 7.02 ± 3.66 in August and 6.96 ± 2.95 in May. It was surprising they were so similar when one considers that densities of the copepods were about an order of magnitude greater in August than in May. This suggests the background density of copepods is related to patch density, for even though the densities of copepods were markedly different in the 2 mo, the ratio between the density in the patches and in the background remained essentially the same.

The zooplankton composition of samples taken on a station did not change during the 4 min needed to make the two tows. The only exceptions were *Mysis mixta* in August and *S. elegans* biomass in the May samples. Evidently the zooplankton patches sampled on each station were too large to be affected by the degree of horizontal water transport occurring within 4 min or the few meters' variation in the boat's position during the first and second tow.

Few sample components in either month showed a significant difference between stations. This suggests that "patch" diameter for most of the zooplankton components was at least as great as the 1.6 km found by Platt et al. (1970).

Sampling at 6-h intervals over a tidal cycle of 12 h gave as good an estimate of zooplankton numbers and biomass as sampling at 2-h intervals over 12 h. If only one sample or a short series of samples were taken on a station, say within 15 min, large overestimates or underestimates of the zooplankton sample could occur. An example of this would be if a zooplankton sample were taken on Station A at 0800 on August 12, 1971, and a sample were taken at 0900 on May 24, 1972. The biomass values would be 16 and 15 g 100 m^{-3}, respectively. But if the samples had

been taken at 1400 on August 11 and 1500 on May 23, the values would have been 25 and 10 g 100 m^{-3}, a difference of 150% compared to 7% as found by the first set of samples. The percentage difference between the actual means measured over the 26-h periods was 80%.

The complexity of the periodic fluctuation varied with different species of copepods and the fact that few were correlated with each other suggested that the different species of copepods were reacting to different environmental conditions. The biomass of zooplankton, consisting mainly of copepods, was much less variable than the numbers in the different species, which suggested that the animals may interact in some way as to reduce extremes in density. Strickler and Bal (1973) found that the copepod *Cyclops abyssorum* maintained a living space of about 40 mm in diameter or about 33,500 mm^3, giving a maximum possible density of about 30,000 animals m^{-1}. The minimum living space needed by an individual copepod probably depends on the size of the animal; therefore, as body size increases the density m^{-1} will decrease, but the total biomass m^{-1} may remain the same or decrease at a lower rate. This may explain the more stable values for zooplankton biomass compared to those for the numbers of copepods of individual species making up the biomass. A similar idea was suggested by Sheldon et al. (1972), using a much greater size range of living particles. They showed a decrease in concentration of particles with increased particle diameter in the range of 100–4000 μ, a range that included all the species of copepods found during this study. What environmental factors influenced the density of copepods can only be surmised. Cassie (1960) accounted for the distribution pattern of zooplankton in an area of mixing between oceanic and harbor water in terms of temperature and salinity preferences of the animals. Boyd (1973) found a significant correlation between concentrations of zooplankton with spherical diameter between 0.531 and 2.55 mm and fast temperature fluctuations. Areas where the temperature showed fluctuations over short distances tended to be areas of high animal concentrations. It seems unlikely that major changes in salinity would occur in St. Margaret's Bay since there are no major sources of fresh water. Bathythermograph readings taken at 2-h intervals during the experiments did not show any major changes in the temperature structure of the water over the 26 h. The bathythermograph is a crude instrument for measuring temperature and it is possible that water with a different temperature structure might have been detected, had more sensitive temperature recording equipment been used. Platt et al. (1970) found concentrations of chlorophyll several times as great as the general background concentrations in St. Margaret's Bay. Therefore, it is possible that the copepods in the present experiments were responding to high concentrations of phytoplankton. However, a satisfactory explanation for concentrations of animals will have to wait for more sophisticated field experiments able to measure several environmental parameters simultaneously.

The diurnal pattern shown by *M. mixta* and *S. elegans* samples can be attributed to the strong vertical migration these animals had during the night. Personal observations from a submarine in August 1968 showed *Mysis* and *Sagitta* to be concentrated in a 2-m layer above the bottom. In addition 200-kHz sonar scattering layers formed by *Mysis* migrating from the bottom to the upper water layers have been observed. It is possible that net avoidance by *Mysis* also played a role in reducing the numbers caught in the early evening and morning when sufficient light was present for the animals to see the net. *Mysis mixta*, one of the most vigorous swimmers among the plankton, showed a significant difference in numbers between tows that might have been due to the animals scattering during the first tow, causing the density to be lower in the region during the second tow. Alternatively, animals disturbed by the first tow may have been more wary of the net during the second tow. If these explanations are correct, it would appear that when using the vertical tow sampling method for *Mysis*, the first tow's results are more likely to be representative of the local density than the second or subsequent tows on the same station. Fleminger and Clutter (1965) found the mysid *Metamysidopis elongata* was able to avoid their nets during the daylight much more effectively than during the night, whereas light intensity had no effect on the number of copepods caught.

The avoidance of nets by copepods was discussed by Clutter and Anaraku (1968) and Laval (1974). Laval examined Fleminger and Clutter's data and concluded that a majority of *Acartia clausi* and *Paracalanus parvus* were able to avoid a 45-cm diam net towed at 30 cm s^{-1}. Laval gave a relationship for zooplankton avoidance and net towing speed that showed an exponential decrease in avoidance with net speed and little reduction in avoidance by copepods when the net was towed above 150 cm s^{-1}. Therefore, if net avoidance by copepods occurred in the present study it was likely quite low and consistent with time.

The number of animals caught per unit volume

of water is a function of the mesh size. Sameoto (1971b) found the #20 (76 μ) mesh net retained 2–4 times as many *Pseudocalanus* and up to 10 times as many *Oithona* individuals as the #6 (243 μ) net. But due to the #20 net retaining large amounts of phytoplankton with the zooplankton it was very difficult to get accurate estimates of the biomass and numbers of zooplankton. The general pattern of biomass and number fluctuations would likely have been similar had the finer mesh been used. But one must keep in mind that the values given for the copepods and other similar sized zooplankton organisms are underestimated.

This study has shown a relationship between tidal fluctuations and the composition of zooplankton samples and suggested ways of reducing the variance results from this effect. It is not known if the tidal effect is peculiar to nearshore waters or if it is general along the entire shelf. This can be determined only by further experiments in offshore waters away from the influence of the coast.

Acknowledgments

I would like to thank Drs A. L. Wilson and A. Douglas of the Computing and Applied Statistics Directorate of the Department of the Environment for their suggestions on experimental design and statistics, Dr L. M. Dickie for his helpful suggestions with respect to the manuscript, Messrs L. O. Jaroszynski, L. Rushton, D. Rudderham, and Miss S. Guildford for their very able technical assistance. Without their help this work would not have been possible.

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Two New Species of *Protanypus* Kieffer, with Keys to Nearctic and Palaearctic Species of the Genus (Diptera: Chironomidae)

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SÆTHER, O. A. 1975. Two new species of *Protanypus* Kieffer, with keys to Nearctic and Palaearctic species of the genus (Diptera: Chironomidae). J. Fish. Res. Board Can. 32: 367–388.

Two Nearctic species of *Protanypus* are described: *Protanypus ramosus* n.sp. from imagines, pupae, and fourth-, third-, and second-instar larvae; *Protanypus hamiltoni* n.sp. from male imagines, pupae, and fourth-, third-, and second-instar larvae. The larvae of two other species are described. These are the first published descriptions of Nearctic imagines of the genus.

Keys are given for male imagines, pupae, and known larvae of the Nearctic and Palaearctic species of the genus.

Protanypus ramosus occurs in several oligotrophic and oligohumic to weakly polyhumic lakes in northwestern Ontario, in Algonquin Park, and in the Great Lakes. *Protanypus hamiltoni* is found in three deep and large, oligotrophic lakes in British Columbia and in Great Slave Lake. Both species are bivoltine. *Protanypus* sp. A occurs in Marion Lake, British Columbia, a small oligotrophic lake. *Protanypus* sp. B is found in George Lake, La Cloche Mountain Region, Ontario, an acidified oligotrophic lake.

SÆTHER, O. A. 1975. Two new species of *Protanypus* Kieffer, with keys to Nearctic and Palaearctic species of the genus (Diptera: Chironomidae). J. Fish. Res. Board Can. 32: 367–388.

Nous décrivons deux espèces néarctiques du genre *Protanypus: Protanypus ramosus* n. sp., à partir d'imagos, de pupes et de larves quaternaires, tertiaires et secondaires; *Protanypus hamiltoni* n.sp., à partir d'imagos mâles, de pupes et de larves quaternaires, tertiaires et secondaires. Nous décrivons les larves de deux autres espèces.

Nous donnons des clés d'identification des imagos mâles, des pupes et des larves connues des espèces néarctiques et paléarctiques du genre.

Protanypus ramosus se rencontre dans plusieurs lacs allant d'oligotrophes et oligohumiques à faiblement polyhumiques du nord-ouest de l'Ontario, dans le parc Algonquin et dans les Grands lacs. *Protanypus hamiltoni* se trouve dans trois lacs oligotrophes profonds et étendus de la Colombie-Britannique et dans le Grand lac des Esclaves. Les deux espèces sont bivoltaines. *Protanypus* sp. A se rencontre dans le lac Marion, Colombie-Britannique, petit lac oligotrophe. *Protanypus* sp. B se trouve dans le lac George, lac oligotrophe acidifié de la région des montagnes La Cloche, Ontario.

Received October 21, 1974
Accepted December 10, 1974

Reçu le 21 octobre 1974
Accepté le 10 décembre 1974

As mentioned by Sæther (1973, p. 665) one of the major shortcomings in analysis of Nearctic benthic chironomid samples has been the lack of knowledge of the specific identities of the typologically more important animals. However, the description of several important members of ultra-oligotrophic, moderately oligotrophic, and mesotrophic communities (Sæther 1973, 1975a, b) has made it possible to develop preliminary lists of indicator communities for North America (Sæther 1975c). One of the more important genera is *Protanypus* Kieff. of which two new species and the larva of two others are described in this paper. There are several records of *Protanypus* from North America (Miller 1941, p. 19; Oliver 1963, p. 177; 1964, p. 79; 1968, p. 116; Hamilton 1965, p. 52; 1971, p. 262; Brinkhurst et al. 1968, p. 17; Hiltunen 1969, p. 129; Sæther 1970, p. 6; Sæther and McLean 1972, p. 8). However, only Oliver (1963, p. 177) gives a specific identification, *P. caudatus* Edw., an identification which he regards

Printed in Canada (J3599)
Imprimé au Canada (J3599)

as more doubtful in consequent papers (Oliver 1964, p. 79; 1968, p. 116).

Methods and Morphology

The mounting procedure used is outlined by Sæther (1969, p. 1). The general terminology follows Sæther (1971, 1974) with the following corrections and modifications discussed by Sæther (1975b): Brachiolum is used instead of basal vein (see Hirvenoja 1973), laterosternite IX instead of laterotergite IX, the tarsal "Sinneszapfen" (Sz) of Hirvenoja (1973) is called sensillae chaeticae, and stipes is used instead of cardostipes (see Hansen 1973). Sæther (1971) used the term aedagal lobe for different structures in his fig. 7A and 8B. The term aedagal lobe should be restricted to the lobe more or less distinctly attached to the phallapodeme (Sæther 1971, fig. 7B, D, 8D, G, 9B). Sæther (1971, p. 1249) followed Abul-Nasr (1950), Strenzke (1959), Wensler and Rempel (1962), and van Emden and Hennig (1970) in terming the dorsal or superior volsella the claspette. According to Hirvenoja (1973, p. 27) this volsella is, in chironomids, not homologous with the claspette of the Culicidae and thus should be named differently. In the definition of Smith (1969) the volsellae are the appendages of the gonocoxite and the gonapophysis or both (excluding the gonostylus), whereas the endomeres ("parameres") correspond with parts of gonapophysis IX, sometimes including part of or the whole volsellae. Hirvenoja (1973, p. 24) divides the endomeres ("parameres") into three parts: lateral, intermedian, and median. In *Protanypus*, at least, the intermedian and the lateral endomere clearly seem to correspond with parts of gonapophysis IX and thus fall within the definition given by Smith (1969). The terms intermedian endomere and lateral endomere are thus used here.

Measurements and ratios follow Schlee (1966) with the additions and modifications given by Sæther (1969). In the following descriptions measurements are given as ranges followed by a mean when four or more measurements have been made and again followed by a number in parenthesis giving the number measured (n).

Holotypes, the allotype of *P. ramosus*, and some paratypes have been deposited in the Canadian National Collection (CNC) in Ottawa. Remaining paratypes are deposited at the Illinois Natural History Survey, Urbana, Ill., returned to lender or retained at the collection of the Freshwater Institute, Winnipeg, Man.

The following abbreviations and ratios are used:

Imagines

$$\text{AR (antennal ratio)} = \frac{\text{ultimate flagellomere}}{\text{remaining flagellomeres}}$$

$$\text{BR (bristle ratio)} = \frac{\text{longest seta of } ta_1}{\text{minimum width of } ta_1 \text{ about } \frac{1}{3} \text{ from apex}}$$

$$\text{BV} = \frac{fe + ti + ta_1}{ta_2 \rightarrow ta_5}$$

fe = femur

$$\text{HR (hypopygium ratio)} = \frac{\text{length of gonocoxite}}{\text{length of gonostylus}}$$

$$\text{HV (hypopygium value)} = \frac{\text{length of male}}{\text{length of gonostylus} \times 10}$$

$$\text{LR (leg ratio)} = \frac{ta_1}{ti}$$

MC_u = media to cubitus crossvein

p_{1-3} = legs 1–3

R, R_1, R_{2+3}, R_{4+5} = stem and branches of radius

RM = radius to media crossvein

$$\text{SV} = \frac{fe + ti}{ta_1}$$

ta_{1-5} = tarsi 1–5

ti = tibia

$$\text{VR (Venarum ratio)} = \frac{Cu}{M}$$

Pupae

D-setae = dorsal setae (see Pagast 1947)

L-setae = lateral setae (see Pagast 1947)

Larvae

$$\text{AR (antennal ratio)} = \frac{\text{length of basal antennal segment}}{\text{length of remaining segments}}$$

Protanypus ramosus n.sp.

The male imago is characterized by having about 60–64 temporals including 2–3 preoculars, a VR of about 0.76, R with 19–23 setae, R_1 with 9–12 setae, LR_1 of 0.69–0.74, SV_2 of 3.79–3.88, ta_1 of p_1 with 7–8 pseudospurs, lateral endomere with scales, intermedian endomere with a long apical tooth and 1–2 short and simple lateral teeth, gonostylus with microtrichiae in proximal 0.4, and part of gonocoxite posterior to gonostylus about 0.7 times as long as gonostylus.

The female has 49–79 temporals including 8–12 preoculars, R with 27–31 setae, R_1 with 15–22 setae, LR_1 of 0.60–0.65, SV_2 of 4.00–4.13, ta_1 of p_1 with 9–12 pseudospurs, 55–60 sensillae chaeticae on ta_1 of p_2 and 72–108 on ta_1 of p_3, gonocoxite IX with 17–29 weak setae, and tergite IX with 51–64 setae.

The pupa has a 0.5–0.7 mm long thoracic horn with very fine spinules for most of its length, tergite I with very weak analmedian shagreenation, tergites II–IX with median shagreenation, sternite I with thin spinules occupying posterior ⅓–½, 9–13 dorsal setae on tergites I–VII, and caudal lobe (male) or lobes (female) of segment VIII apically more or less truncated.

The larva has 11–12 labral scales and an AR of about 2.2; apical style of second antennal segment is about twice as long as accessory style and about as long as segment 3 and 4 combined; medioventral appendix of prementum has un-

evenly serrated blades and the widest part is about 2.4–2.5 times as wide as the narrowest part.

MALE IMAGO (N = 2, EXCEPT WHEN STATED OTHERWISE)

Length 6.27–6.78 mm. Wing length 4.06–4.22 mm. Total length/wing length: 1.54–1.61. Wing length/profemur: 2.51–2.59.

Head — AR = 2.52 (1). Temporal setae 60–64. Postorbitals 12–18, divided into two groups each with 6–9 setae; outer verticals 8–9, forming a continuous group with upper group of postorbitals; inner verticals 35–37; preoculars 2–3. Clypeus without setae. Cibarial pump, tentorium, and stipes as in Fig. 1A. Tentorium 310 μ(1) long, maximum width 66 μ(1), width anterior of posterior tentorial pit 21 μ(1), distance from apex to posterior tentorial pit 34 μ(1). Stipes 310 μ long, the two apparently fused medially. Two vestigial ocelli 100 μ(1) apart. Palp lengths in microns: 80, 124–130, 204–232, 220–244, 304–350. Sensilla coeloconicae of third segment large, conspicuous, but without any sensillae chaeticae.

Thorax (Fig. 1B) — Antepronotum with 7–11 median setae and 42–44 lateral setae. Medium anepisternum II with 2–4 setae, posterior anepisternum II with 14–17, epimeron II with 12, preepisternum with 30–35 setae. Dorsocentrals 28; acrostichals 30–35, up to 90 μ long; prealars 19 with a group of 14–15 posterior, 3–4 anterior, and 1 intermediate; supraalars 3, parascutellars 1–2. Scutellum with 40–49 setae.

Wing — VR = 0.76. Extended part of costa 75–80 μ long. MC_u 50–55 μ from RM. Brachiolum with 5–6 setae, R with 19–23, R_1 with 9–12, R_{4+5} with 3 setae. Squama with 72–73 setae. Sensillae campaniformes 17–18 at base of brachiolum, 4–5 below setae of brachiolum, 11–12 at apex of brachiolum, five in basal 260–280 μ of subcosta, one 110–200 μ from base on R_1, and one dorsal and one ventral 60–80 μ and 70–110 μ, respectively, from base on R_{2+3}.

Legs — Spur of front tibia (Fig. 2B) 132–142 μ long, spurs of middle tibia (Fig. 2C) 92–116 and 90–107 μ long, of hind tibia (Fig. 2D) 122–165 μ and 74–90 μ. Width at apex of front tibia 92–100 μ, of middle tibia 80–94 μ, of hind tibia 104–110 μ. No true comb present, but heavier spiniform setae in apical of hind tibia grade over in an irregular apical row. Front leg with 7–8 tarsal pseudospurs on ta_1, 2–3 on ta_2, 1–2 on ta_3. Middle leg with 13–16 pseudospurs on ta_1, 5 on ta_2, 2–3 on ta_3. Hind leg with 21–23 pseudospurs on ta_1, 9 on ta_2, 3–4 on ta_3. Pseudospurs 50–60 μ long on ta_1, 40–60 μ on ta_2, and 35–50 μ on ta_3. Lengths (in microns) and proportions of legs:

	fe	ti	ta_1
p_1	1616–1626	1872–2079	1380–1438
p_2	1695–1852	1813–2049	926–1005
p_3	1961–2039	2306–2522	1419–1498
	ta_2	ta_3	ta_4
p_1	662–668	429–442	245–258
p_2	466–478	307–319	202–209
p_3	675–748	393–429	258–276
	ta_5	LR	BV
p_1	159–209	0.69–0.74	3.23–3.29
p_2	165–172	0.49–0.51	3.89–4.16
p_3	221–227	0.59–0.62	3.62–3.66
	SV	BR	
p_1	2.53–2.58	3.25–3.33	
p_2	3.79–3.88	3.05–3.18	
p_3	3.01–3.04	3.04–3.25	

Hypopygium (Fig. 1C) — Ninth tergum with 32–36 setae, laterosternites IX each with 10 setae. Phallapodeme 240–250 μ long. Lateral endomere with scales, 190–206 μ long. Intermedian endomere (Fig. 1D, E) 150–220 μ long, with long apical tooth (60–90 μ long) and 1–2 shorter lateral teeth (16–30 μ). Gonocoxite 440 μ long, with a 190 μ long apical projection. Gonostylus 264–270 μ long, with microtrichiae in proximal 0.41–0.44. HR without projection = 1.63–1.67; HR with projection = 2.33–2.39; HV = 2.32–2.57.

FEMALE IMAGO (N = 5, EXCEPT WHEN STATED OTHERWISE)

Length 6.70–7.39, 6.96 mm. Wing length 4.77–5.12, 4.95 mm(4). Total length/wing length: 1.37–1.44, 1.42(4). Wing length/profemur: 2.85–2.97, 2.93(4).

Head — AR = 0.21–0.26, 0.23(4). Scapus with 12–17,14(4) setae. Flagellomeres 13, 120–164,136 μ (4) long. Temporal setae 49–79,63(4). Postorbitals divided into two groups with 8–10,10(4) setae in lower anterior group and 7–9,8(4) in upper posterior group; outer verticals 6–17,12(4); inner verticals 18–32,24(4); preoculars 8–12,10(4). Clypeus without setae. Coronal suture absent. Tentorium (Fig. 2A), 345–360,351 μ(4) long; maximum width 66–80,75 μ(4); width anterior of posterior tentorial pit 28–40 μ(3), distance from apex to posterior tentorial pit 14–22

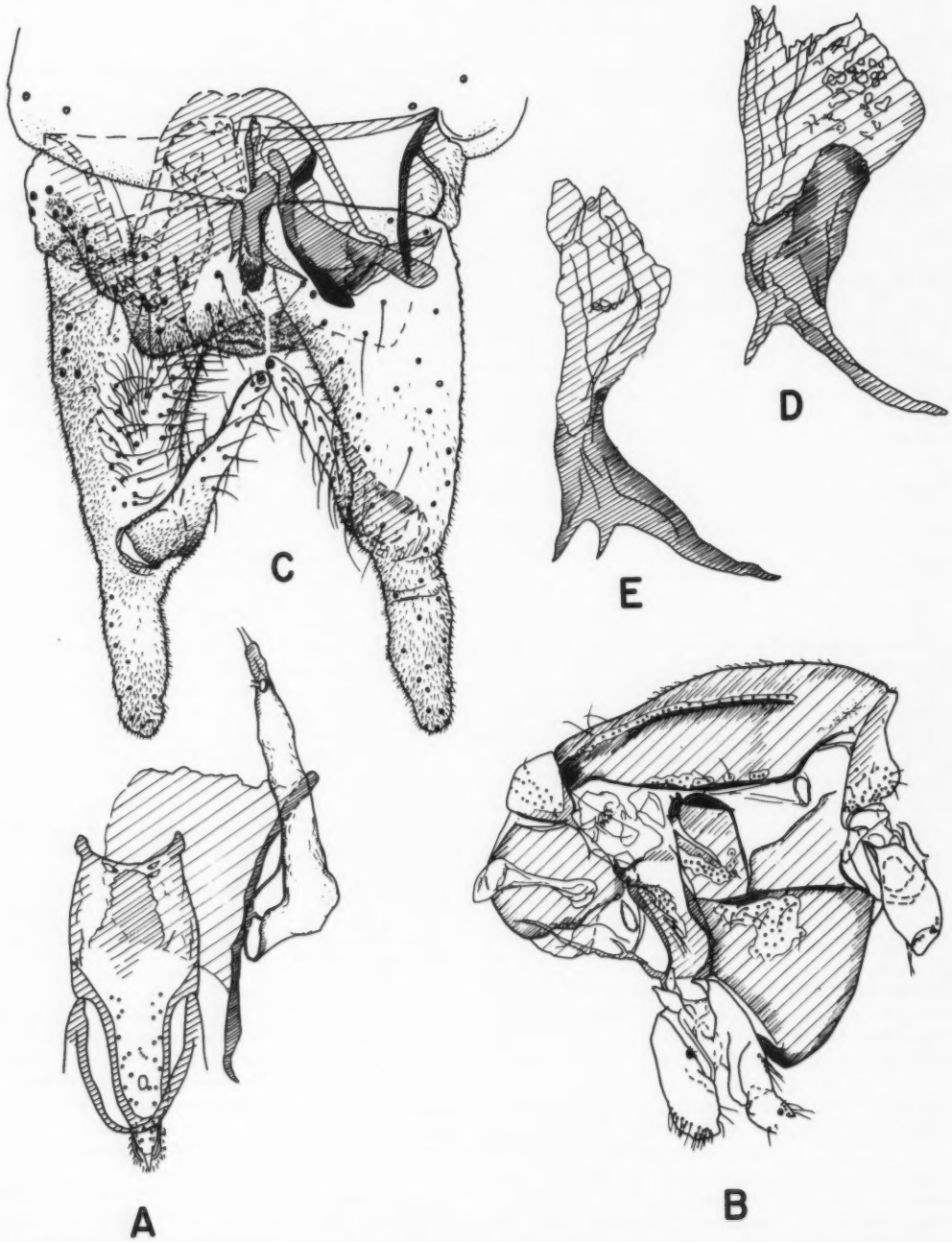

FIG. 1. *Protanypus ramosus* n.sp., male. A, cibarial pump, labial lonchus, tentorium, and stipes; B, thorax; C, hypopygium; D, E, intermedian endomere on holotype from Kenora (D) and paratype from Costello Lake (E).

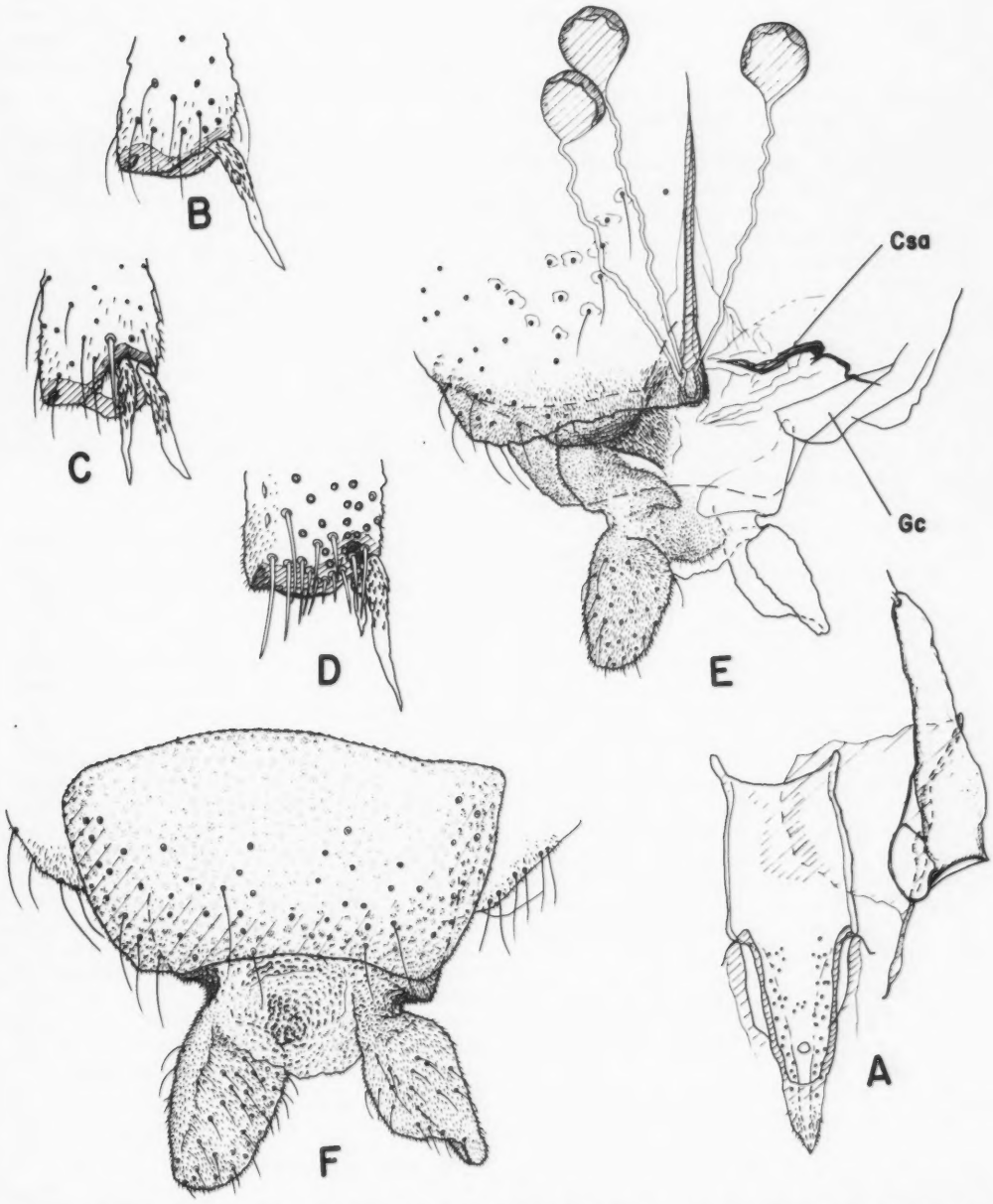

FIG. 2. *Protanypus ramosus* n.sp. A, cibarial pump, labial lonchus, tentorium, and stipes of female; B, C, D, apex of front, median, and hind tibia, respectively, of male; E, F, female genitalia, ventral (E) and dorsal (F) view. (Csa: coxosternapodeme; Gc: gonocoxite (IX).)

μ(3). Palp lengths in microns (n = 4): 88–98,95; 130–142,136; 222–236,231; 210–266,239; 310–362,346.

Thorax — Antepronotum with 64–107,75 setae. Median anepisternum II with 5–8,6 setae; posterior anepisternum II with 13–20,16; epimeron II with 10–20,14; preepisternum with 32–40,35. Dorsocentrals 32–41,37; acrostichals 28–41,33; prealars 33–49,39, with 6–10 anterior ones more or less separate from posterior ones; supraalars 2–4,3; parascutellars 1–4,3. Scutellum with 58–77,68 setae.

Wing (n = 4) — VR = 0.83–0.84, 0.83. Extended part of costa 40–60, 51 μ. Brachiolum with 7–9,8 setae; R with 27–31,29; R_1 with 15–22,19; R_{2+3} with 1 seta in two of four specimens; R_{4+5} with 8–20, 15 setae. Squama with 96–117,105 setae. Sensillae campaniformes 18–23,21 at base of brachiolum; 4 below setae of brachiolum; 8–9,9 at apex of brachiolum; 4 in basal 210–300,255 μ of subcosta; one 200–280,245 μ from base on R_1; 1–2,1.5 dorsal and ventral 32–140,97 μ from base on R_{2+3}.

Legs — Spur of front tibia 119–128, 124 μ long; spurs of middle tibia 89–100,95 μ and 86–100,94 μ; of hind tibia 126–140,131 μ and 98–106,101 μ. Width at apex of front tibia 96–102,100 μ; of middle tibia 104–110,108 μ; of hind tibia 110–126,119 μ. Front leg with 9–12,10 tarsal pseudospurs on ta_1; 2 on ta_2; 0–2 on ta_3. Middle leg with 14–19,16 pseudospurs on ta_1; 5–6 on ta_2; 2 on ta_3. Hind leg with 23–26,24 pseudospurs on ta_1; 7–10,8 on ta_2; 2–3 on ta_3. Pseudospurs 45–60 μ long on ta_1, 40–50 μ on ta_2, 35–50 on ta_3. Sensillae chaeticae 55–60,58(4) in basal ⅛ to ¼ on p_2; 72–108,88(4) in basal ⅛ to ¼ on p_3. Lengths (in microns) and proportions of legs:

	fe	ti	ta_1
p_1	1596–1754, 1673	2040–2237, 2109	1252–1380, 1311
p_2	1813–1971, 1892	1941–2128, 2028	926–1005, 963
p_3	2030–2188, 2125	2523–2720, 2627	1390–1518, 1467
	ta_2	ta_3	ta_4
p_1	352–626, 594	393–442, 417	258–294, 277
p_2	423–466, 445	331–356, 343	227–239, 233
p_3	699–761, 720	443–466, 445	282–313, 296
	ta_5	LR	BV
p_1	215–245, 231	0.60–0.65, 0.62	3.19–3.50, 3.35
p_2	202–227, 217	0.47–0.49, 0.48	3.83–4.02, 3.94
p_3	233–258, 247	0.55–0.57, 0.56	3.56–3.77, 3.64
	SV	BR	
p_1	2.76–2.98, 2.88	2.03–2.22, 2.12	
p_2	4.00–4.13, 4.07	1.67–2.10, 1.91	
p_3	3.15–3.31, 3.24	2.00–2.33, 2.16	

Genitalia (Fig. 2E, F) — Gonocoxite with 17–19,22 weak setae. Tergite IX undivided, with 51–64,57(4) setae. Cercus 200–210,205 μ long. Three ovoid seminal capsules each 110–134,119 μ long and 93–108,103 μ wide.

PUPA (N = 4, EXCEPT WHEN STATED OTHERWISE)

Length 8.02–10.80,9.38 mm. Length of thoracic horn/length of anal macrosetae: 1.37–1.44(2).

Cephalothorax — Thoracic horn (Fig. 3A) 546–687 μ(3) long, 56–80 μ(3) wide, covered with faint spinules for most of its length, mostly with a proximal swelling. Frontal tubercle (Fig. 3B, C) 100 μ(1) high with the wider base an additional 100 μ(1) high, tubercle 70–80 μ(2) wide, base 140–150 μ(2) wide; apical setae 527–552 μ(3) long. Anterior thoracic tubercle (Fig. 3B, C) 70–90 μ(2) high, 66–70 μ(2) wide, with a 331–491, 428 μ(4) long apical seta.

Abdomen — Tergite I with weak and sparse median spinules in posterior corners of polygons of reticulation, remaining tergites fully shagreenated except on lateral margins. Sternite I (Fig. 3D) with shagreenation in posterior ⅓ to ½ consisting of thin 5–6 μ long spinules, sternite II with up to 10 μ long spinules, remaining sternites with spinules of same length as on tergites (up to 7 μ), sternites II–V fully shagreenated except laterally, sternite VI–VIII with sparse median shagreenation. Tergites I–VII with 9–13 pairs of dorsal setae (D-setae), tergite VIII with 6–7 setae. Segments I–VIII with 3 L-setae, none filamentous on I, 2 filamentous on II–IV, all 3 filamentous on V–VIII. Caudal lobe of segment VIII in male (Fig. 3F) 399–515 μ(3) long, usually apically truncate, 0.53–0.67(3) times as long as anal lobe. Caudal lobes of segment VIII in female (Fig. 3E) 711 μ(1) long, ending 552 μ(1) posterior of tergite VIII. Anal lobe 662–853, 761 μ(4) long; with 7–13,10(4) setae; 4–5 of them on strong tubercles; 0–3,1 on weaker tubercles; setae of tubercles 343–417 μ(3) long, shortest setae 221–307 μ(3) long.

FOURTH-INSTAR LARVA (N = 2, EXCEPT WHEN STATED OTHERWISE)

Length 7.84–10.45 mm. Head capsule length 0.77–0.82 mm.

Head — Antenna as in Fig. 4A. Lengths of antennal segments in microns: 80–98, 25–28, 4,

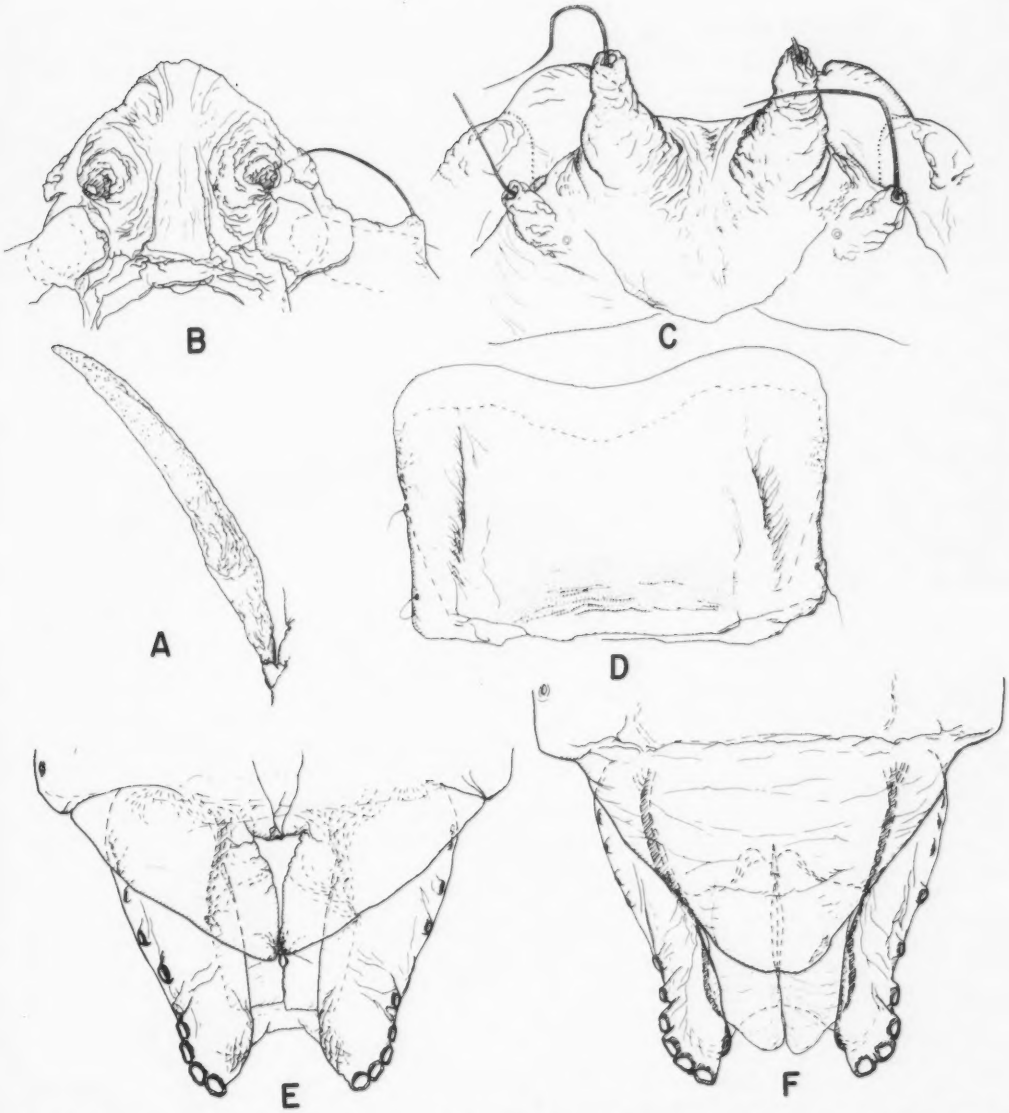

FIG. 3. *Protanypus ramosus* n.sp., pupa. A, thoracic horn; B, C, frontal plate and anterior thoracic tubercle, different views; D, sternite I; E, F, caudal lobes of segment VIII and anal lobe of female (E) and male (F).

8–9. AR = 2.22. Basal antennal segment 28–30 μ wide, 2.86–3.27 as long as wide, distance from base to annular organ 12–15 μ, to basal seta 22 μ(1), blade at apex 30–33 μ long, accessory blade 30–33 μ long. Apical style of second segment 12–13 μ long, accessory style 6 μ. Labrum and palatum as in Fig. 4B, labrum with 11–12 scales. Premandible 98 μ(1) long. Maxilla as in Fig. 4C, D. Medioventral appendix of prementum (Fig. 4E) 122–140 μ long, 76–92 μ wide, minimum width at base 31–37 μ, maximum width/basal minimum width: 2.45–2.47. Supporting endoskeleton of appendix 132–152 μ wide, 1.65–1.69 times as wide as the 80–90 μ median tooth

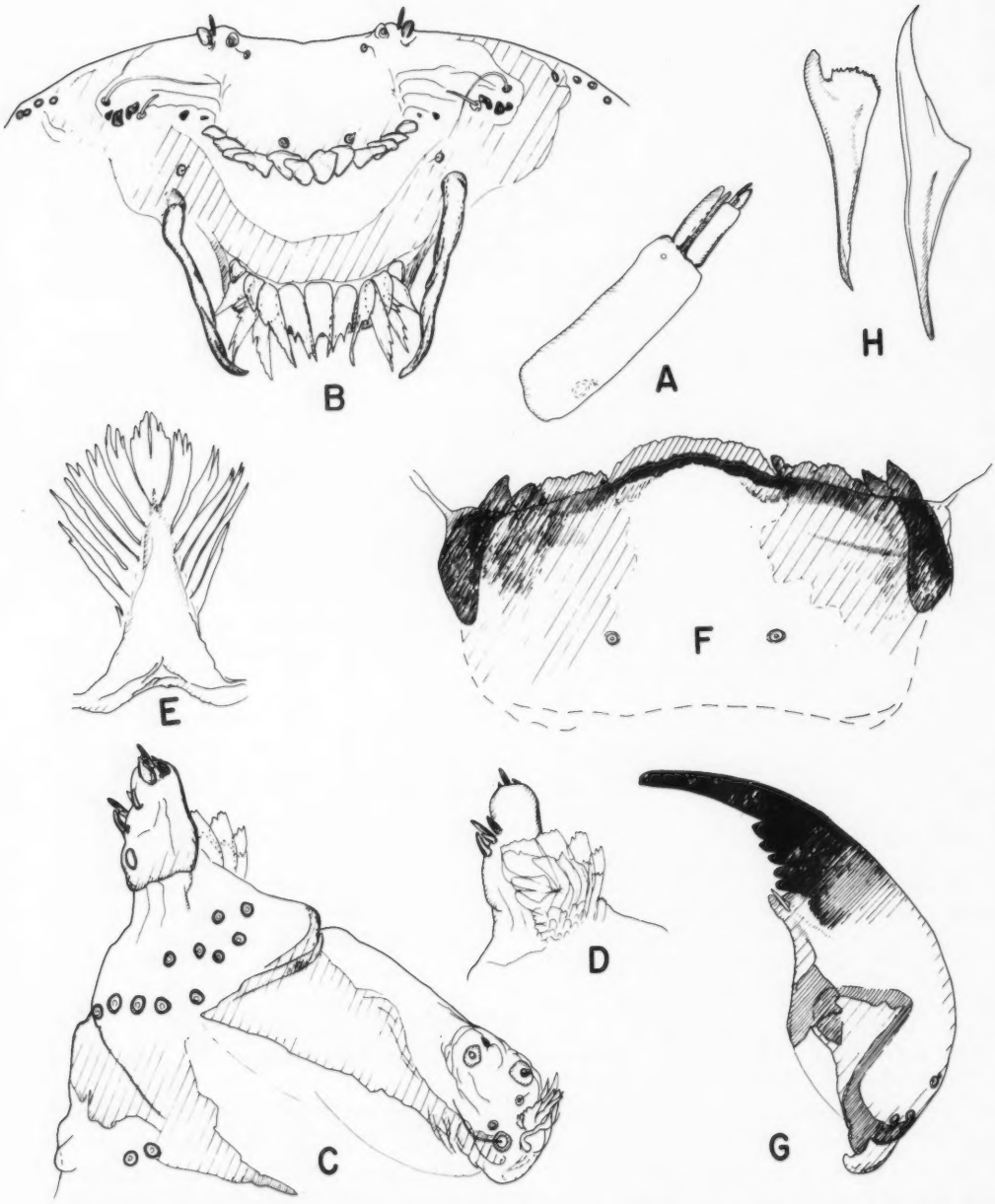

FIG. 4. *Protanypus ramosus* n.sp., larva. A, antenna; B, labrum and palatum; C, D, maxilla and maxillary palp, ventral (C) and dorsal view (D); E, medioventral appendix of prementum; F, mentum; G, mandible; H, claws of posterior parapods.

of mentum (Fig. 4F). Mandible (Fig. 4G) 312 μ(1) long, apical tooth 79–92 μ long. Postmentum (mentum plus submentum) 302–348 μ long.

Abdomen — Procercus 114–134 μ high, 36–42 μ wide, with 6 apical anal setae and 2 weak lateral setae. Posterior parapods 613 μ(1) long with two kinds of apical claws (Fig. 4H).

THIRD-INSTAR LARVA (N = 6, EXCEPT WHEN STATED OTHERWISE)

Length 4.40–7.19, 5.98 mm. Head capsule length 0.45–0.50, 0.48 mm.

Head — Lengths of antennal segments in microns: 49–52,50; 20–27,23; 4; 6–8,7. AR = 1.23–1.58, 1.46. Basal antennal segment 19–20,20 μ wide; 2.45–2.63,2.58 times as long as wide; distance from base to annular organ 6–14,10 μ(5); to basal seta 13–18 μ(2); blade at apex 25–30,28 μ long; accessory blade 25–30, 28 μ. Apical style of second segment 9–11, 10 μ long; accessory style 4–6,5 μ long. Labrum with 8–10,9(5) scales. Premandible 74–81,78 μ(4) long. Medioventral appendix of prementum 78–87,81 μ long; 45–54,49 μ wide; minimum width 19–21,20 μ; maximum width/minimum width: 2.36–2.70,2.45. Supporting endoskeleton of appendix 90–93, 92 μ(5) wide; 1.48–1.80,1.65(4) times as wide as the 50–63,56 μ(4) wide median tooth of mentum. Mandible 200–228,211 μ(4) long; apical tooth 50–56,53 μ(4) long. Postmentum 216–242,233 μ long.

Abdomen — Procercus 72–80, 75 μ high; 25–44,33 μ wide. Anal tubules 116–180 μ(3) long, 38–60 μ(3) wide at base. Posterior parapods 319–393, 358 μ(5) long.

SECOND-INSTAR LARVA (N = 1)

Head capsule length 0.29 mm.

Head — Lengths of antennal segments in microns: 20,18,3,5. AR = 0.80. Basal antennal segment 11 μ wide, 1.82 times as long as wide, blade at apex 22 μ long, accessory blade 22 μ long. Apical style of second segment 6 μ long, accessory style 3 μ long. Labrum with 2 scales. Premandible 44 μ long. Mandible 122 μ long. Postmentum 142 μ long.

Material Examined

HOLOTYPE: male with exuvia, emergence trap, Lake 122, Freshwater Institute Experimental Lakes Area (ELA), Kenora, Ont., 1/10/68, S. S. *Chang* (CNC No. 13403). ALLOTYPE: female, Costello Lake, Algonquin Park, Ont. 17/5/65, D. W. Webb. PARATYPES: 1 male, 4 females, same data as allotype; 2 exuviae, Lake 122, ELA, Kenora, Ont., 7/5 and 14/5/68, S. S. *Chang*. OTHER MATERIAL: pupa with larval exuvia, depth 43 m, Georgian Bay, Lake Huron, 23/8/64, Great Lakes Institute, University of Toronto; 1 fourth-instar larva, depth 26 m, Lake 305, ELA, Kenora, Ont. 2/5/69, A. L. Hamilton and G. P. McRae; 2 third- and 1 second-instar larvae, depth 9–12 m, Lake 239, ELA, Kenora, Ont., 1/5/69, A. L. Hamilton and G. P. McRae; 2 third-instar larvae, depth 1.8 m, Lake Mad Dog, ELA, Kenora, Ont., 6/5/69, A. L. Hamilton and G. P. McRae; 1 third-instar larva, depth 46 m, Hillock Lake, ELA, Kenora, Ont. 6/5/69, A. L. Hamilton and G. P. McRae; 2 third-instar larvae, depth 4 m, South Indian Lake, Man. 12/7/72, A. L. Hamilton.

Remarks

The species most closely resembling *P. ramosus* n.sp is *P. hamiltoni* n.sp. (see p. 368).

Protanypus hamiltoni n.sp.

Protanypus sp. A pro parte, Hamilton 1965 part II; 52 (male, pupa, larva)
Protanypus near *morio* Sæther 1971; fig. 3E, 4D, 8B, 10D (male, larva).

The male imago is characterized by having about 31–40 temporals; lacking preoculars; a VR of 0.82–0.88; R with 29–35 setae; R_1 with 20–26 setae; LR_1 of 0.66–0.70; SV_2 of 4.00–4.06; ta_1 of p_1 with only 2 apical pseudospurs; lateral endomere with scales; intermedian endomere with a short apical tooth and 1–2 short and 1 long, simple to four-pronged lateral teeth; gonostylus with microtrichiae in apical 0.8; and part of gonocoxite posterior to gonostylus about 0.8 times as long as gonostylus.

The pupa has a 0.5–0.7 mm long thoracic horn without spinules; tergite I with weak analmedian shagreenation; tergites II–IX with median shagreenation; sternite I with long, thin spinules occupying posterior ½–⅔; 9–13 dorsal setae on tergites I–VII; and caudal lobe (male) or lobes (female) of segment VIII apically rounded.

The larva has 10–12 labral scales and AR of

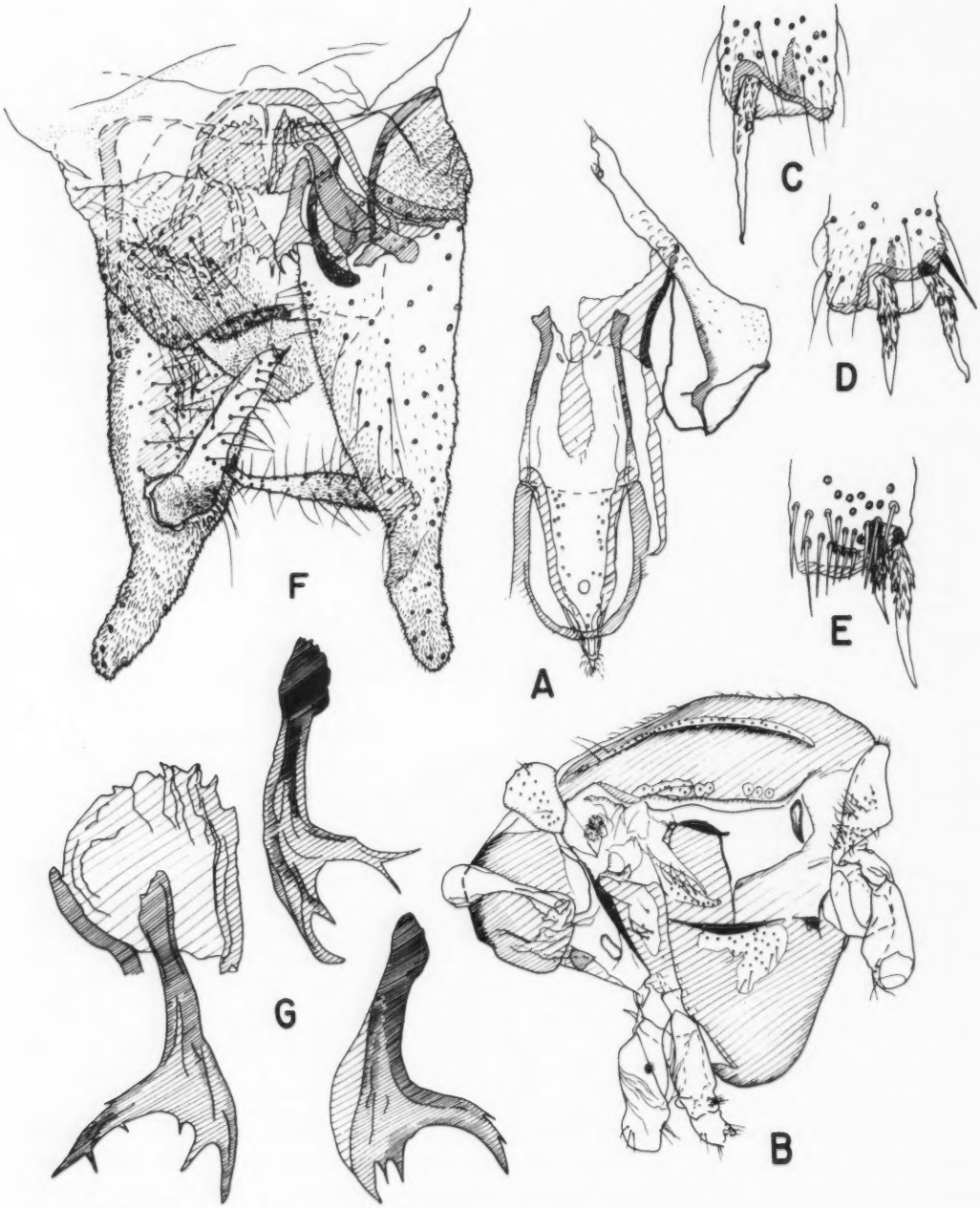

FIG. 5. *Protanypus hamiltoni* n.sp., male. A, cibarial pump, labial lonchus, tentorium, and stipes; B, thorax; C, D, E, apex of front, median, and hind tibia, respectively; F, hypopygium; G variations of intermedian endomere.

2.3–2.7; apical style of second antennal segment is about twice as long as accessory style and about as long as segment 3 and 4 combined; medioventral appendix of prementum has unevenly serrated blades and is 1.9–2.2 times as wide at the widest as at the most narrow point.

MALE IMAGO (N = 6, EXCEPT WHEN STATED OTHERWISE)

Length 6.62–7.19 mm (3). Wing length 3.75–4.53, 4.22 mm (7). Total length/wing length: 1.58–1.67(3). Wing length/profemur: 2.59–2.68, 2.66(4).

Head — AR = 2.30–2.72,2.58(8). Temporal setae 31–40,37. Postorbitals divided into two groups with 6–11,8 setae in lower anterior group and 8–14,11 in upper posterior group, outer verticals 6–9,8; inner verticals 8–12,10. Clypeus without setae. Cibarial pump, tentorium, and stipes as in Fig. 5A. Tentorium 330–380,354 μ(5) long; maximum width 66–78,70 μ(5); width anterior of posterior tentorial pit 18–21,20 μ(5); distance from apex to posterior tentorial pit 23–40,32 μ (5). Palp lengths in microns (n = 7): 68–85, 79; 120–148,136; 210–240,223; 214–272,255; 275–374,335. Sensillae coeloconicae of third segment large, conspicuous, but without any sensillae chaeticae.

Thorax (Fig. 5B) — Antepronotum with 8–12, 10 median setae and 30–49,38 lateral setae. Median anepisternum II with 1–2,1.5 setae; posterior anepisternum II with 13–17,15 setae; epimeron II with 4–9,6(5) setae; preepisternum with 15–30,25 setae. Dorsocentrals 20–26,22(5); acrostichals 22–31,26(5), up to 90 μ long; prealars 14–22,19(5) with a group of 12–19 posterior and 2–4 anterior; supraalars 4–8(3); parascutellars 3–5,4(5). Scutellum with 38–50,43 setae.

Wing — VR = 0.82–0.88,0.85. Extended part of costa 88–120,103 μ long. MC_u 50–60,56 μ(5) from RM. Brachiolum with 5–8,6 setae; R with 29–35,32(7); R_1 with 20–26,23(7); R_{4+5} with 3–8,6(7) setae. Squama with 71–88,77(7) setae. Sensillae campaniformes 19–24,22 at base of brachiolum; 4–5,5 below setae of brachiolum; 10–11,11 at apex of brachiolum; 4–5,4 in basal 240–400,253 μ of subcosta; one 200–450,292 μ from base on R_1; one dorsal and one ventral 95–140,118 μ and 125–170,155 μ from base on R_{2+3}.

Legs (n = 4) — Spur of front tibia (Fig. 5C) 130–142,135 μ long; spurs of middle tibia (Fig. 5D) 101–113,105 μ and 86–102,94 μ; of hind tibia (Fig. 5E) 134–136,135 μ and 84–88,86 μ. No true comb present, but heavier spiniform setae in apical $\frac{1}{10}$ of hind tibia grade over in an irregular apical row. Front leg with 2 tarsal pseudospurs on ta_1; 1–2,1.8 on ta_2; 0–2,1 on ta_3. Middle leg with 12–16,14 pseudospurs on ta_1; 4–8,5 on ta_2; 1–2,1.8 on ta_3. Hind leg with 16–20,19 pseudospurs on ta_1; 5–10,8 on ta_2; 1–2,1.5 on ta_3. Pseudospurs 40–60 μ long on ta_1, 35–55 μ on ta_2, and 35–47 μ on ta_3. Lengths (in microns) and proportions of legs:

	fe	ti	ta_1
p_1	1557–1695, 1611	1912–2148, 2015	1311–1429, 1355
p_2	1734–1911, 1793	1863–2174, 1985	887–1005, 936
p_3	1872–1991, 1929	2384–2503, 2444	1399–1439, 1419

	ta_2	ta_3	ta_4
p_1	662–687, 675	429–466, 454	251–276, 261
p_2	478–527, 506	319–368, 347	196–215, 207
p_3	711–779, 744	380–405, 390	245–258, 253

	ta_5	LR	BV
p_1	202–215, 209	0.66–0.70, 0.68	2.99–3.29, 3.11
p_2	190–196, 193	0.46–0.48, 0.47	3.59–3.95, 3.77
p_3	221–233, 227	0.57–0.59, 0.58	3.57–3.60, 3.59

	SV	BR (n = 3)
p_1	2.59–2.74, 2.68	2.80–3.26
p_2	4.00–4.06, 4.04	2.60–2.95
p_3	3.02–3.17, 3.08	2.90–3.04

Hypopygium (Fig. 5F) — Ninth tergum with 28–32,30(5) setae; laterosternites IX each with 12–15,13(5) setae. Phallapodeme 180–190,187 μ(7) long. Lateral endomere with scales, 152–180,165 μ(7) long. Intermedian endomere (Fig. 5G) 150–170,163 μ(7) long with short apical tooth, 1–2 short lateral teeth and 1 long, single to 4-pronged lateral tooth. Gonocoxite 440–474,455 μ long; with a 200–214,205 μ long apical projection. Gonostylus 254–266,260 μ long; with microtrichiae in proximal 0.81–0.92,0.84. HR without projection = 1.67–1.81,1.75; HR with projection = 2.44–2.62,2.54; HV = 2.51–2.72(3)

PUPA (N = 10, EXCEPT WHEN STATED OTHERWISE)

Length 9.26–10.54,9.85 mm. Length of thoracic horn/length of anal macrosetae: 1.33–1.97,1.56.

Cephalothorax — Thoracic horn (Fig. 6A) 491–675,565 μ long; 62–98,75 μ wide; void of spinules. Frontal tubercle (Fig. 6 B, C), 100–140,116 μ(5) high with an additional 175–240, 199 μ(5) high base; tubercle 66–98,78 μ(9) wide; base 140–230,170 μ(5) wide; apical setae 319–480,412 μ(7) long. Anterior thoracic tubercle (Fig. 6 B, C) 56–86,61 μ(9) high; 56–80,68 μ(9) wide; apical seta 245–440,341 μ(8) long.

Abdomen — Tergite I with weak and sparse median spinules in posterior corners of polygons of reticulation, remaining tergites fully shagreenated except on lateral margins. Sternite I (Fig. 6D) with shagreenation in posterior ⅓–⅔ consisting of thin spinules up to 12 μ long, sternite II with similar long spinules, remaining sternites with spinules of approximately same length as on tergites (up to 7 μ), sternites II–V fully shagreenated except laterally, sternites VI–VIII with sparse median shagreenation. Tergites I–VII with 9–13 D-setae, tergite VIII with 6–7 D-setae. Segments I–VIII with 3 L-setae, none filamentous on I, 2 filamentous on II–IV, all 3 filamentous on V–VIII. Caudal lobe of segment VIII in male (Fig. 6F) 478–540,502 μ(6) long, apically rounded, 0.68–0.74,0.70(6) times as long as anal lobe. Caudal lobes of segment VIII in female (Fig. 6E) 589–613,607 μ(4) long, ending 368–411,391 μ(4) posterior of tergite VIII. Anal lobe 687–797,730 μ long; with 7–9,8 setae; 4–6 of them on strong tubercles, 0–2 on weaker tubercles; setae on tubercles 319–399,365 μ long, shortest setae 172–282,247 μ long.

FOURTH-INSTAR LARVA (N = 5 EXCEPT WHEN STATED OTHERWISE)

Length 6.70–12.22,10.35 mm. Head capsule length 0.74–0.79,0.78 mm.

Head — Antenna as in Fig. 7A. Lengths of antennal segments in microns: 88–104,97; 23–27, 25; 3–5,4; 6–9,8. AR = 2.32–2.65,2.46. Basal antennal segment 30–31,30 μ wide; 2.93–3.47, 3.23 times as long as wide; distance from base to annular organ 16–20,18 μ(4); to basal setae 21–24 μ(3); to mark of distal setae 72–92 μ(3); blade at apex 30–34,32 μ long; accessory blade 31–34,32 μ long. Apical style of second segment 11–14,12 μ long; accessory style 5–8,7 μ long. Labrum and palatum as in Fig. 7B, labrum with 10–12,11 scales. Premandible 120–144,128 μ long. Maxilla as in Fig. 7 C, D. Medioventral appendix of prementum (Fig. 7E) 128–144,135 μ long; 76–90,82 μ wide, minimum width at base 37–42,40 μ; maximum width/basal minimum width: 1.90–2.18,2.05. Supporting endoskeleton of appendix 140–152,136 μ wide; 1.33–1.60,1.50 times as wide as the 93–110,98 μ wide median tooth of mentum (Fig. 7F). Mandible (Fig. 7G) 300–344,318 μ long; apical tooth 86–92,87 μ long. Postmentum 354–400,374 μ long.

Abdomen — Procercus 114–136,124 μ high; 38–46,41 μ wide: with 6 apical anal setae 368–466,414 μ(4) long and 2 weak lateral setae. Supraanal setae 80 μ(1) long. Anal tubules triangular; 190–220,202 μ long; 70–104,82 μ wide at base. Posterior parapods 540–638,589 μ(4) long with two kinds of apical claws (Fig. 7H).

THIRD-INSTAR LARVA (N = 1)

Head capsule length 0.47 mm.

Head — Lengths of antennal segments in microns: 42,21,3,7. AR = 1.27. Basal antennal segment 19 μ wide, 2.21 times as long as wide; distance from base to annular organ 2 μ, to basal seta 6 μ, to distal mark of seta 32 μ; blade and accessory blade at apex both 27 μ long. Apical style of second segment 10 μ long, accessory style 5 μ long. Labrum with 8 scales. Premandible 75 μ long. Medioventral appendix of prementum 79 μ long, 50 μ wide; minimum width at base 26 μ, maximum width/minimum width: 1.92. Supporting endoskeleton of appendix 86 μ wide, 1.51 times as wide as the 57 μ wide median tooth of mentum. Mandible 232 μ long, apical tooth 47 μ long. Postmentum 225 μ long.

SECOND-INSTAR LARVA (N = 1)

Length 3.65 mm. Head capsule length 0.25 mm.

Head — Lengths of antennal segments in microns: 12, 17, 2, 5. AR = 0.46. Basal antennal segment 12 μ wide, as long as wide; blade and accessory blade at apex both 21 μ long. Apical style of second segment 6 μ long, accessory style 2 μ long. Mandible 106 μ long, apical tooth 30 μ long. Postmentum 150 μ long.

Abdomen — Procercus 48 μ high, 24 μ wide. Anal tubercles 80 μ long, 40 μ wide at base.

Material Examined

HOLOTYPE: male, Babine Lake, B.C., 18/5/65, A. L. Hamilton (CNC No. 13402). PARATYPES: 6 males, 1 mature pupa, 13 pupal exuviae, 2 larvae (depth 3.5 m), same data as holotype; 1 male, Hay River, N.W.T., 10/9/32, O. Bryant; 2 pupal exuviae, Westbank, Okanagan Lake, B.C., 9/9/69, J. F. Flannagan and O. A. Sæther; 14 larvae depths 13–117 m, Okanagan Lake, B.C., 8-10/9/69, J. F. Flannagan and O. A. Sæther; 4 larvae, depths 37–100 m, Kalamalka Lake, B.C., 11/6/71, J. F. Flannagan and O. A. Sæther.

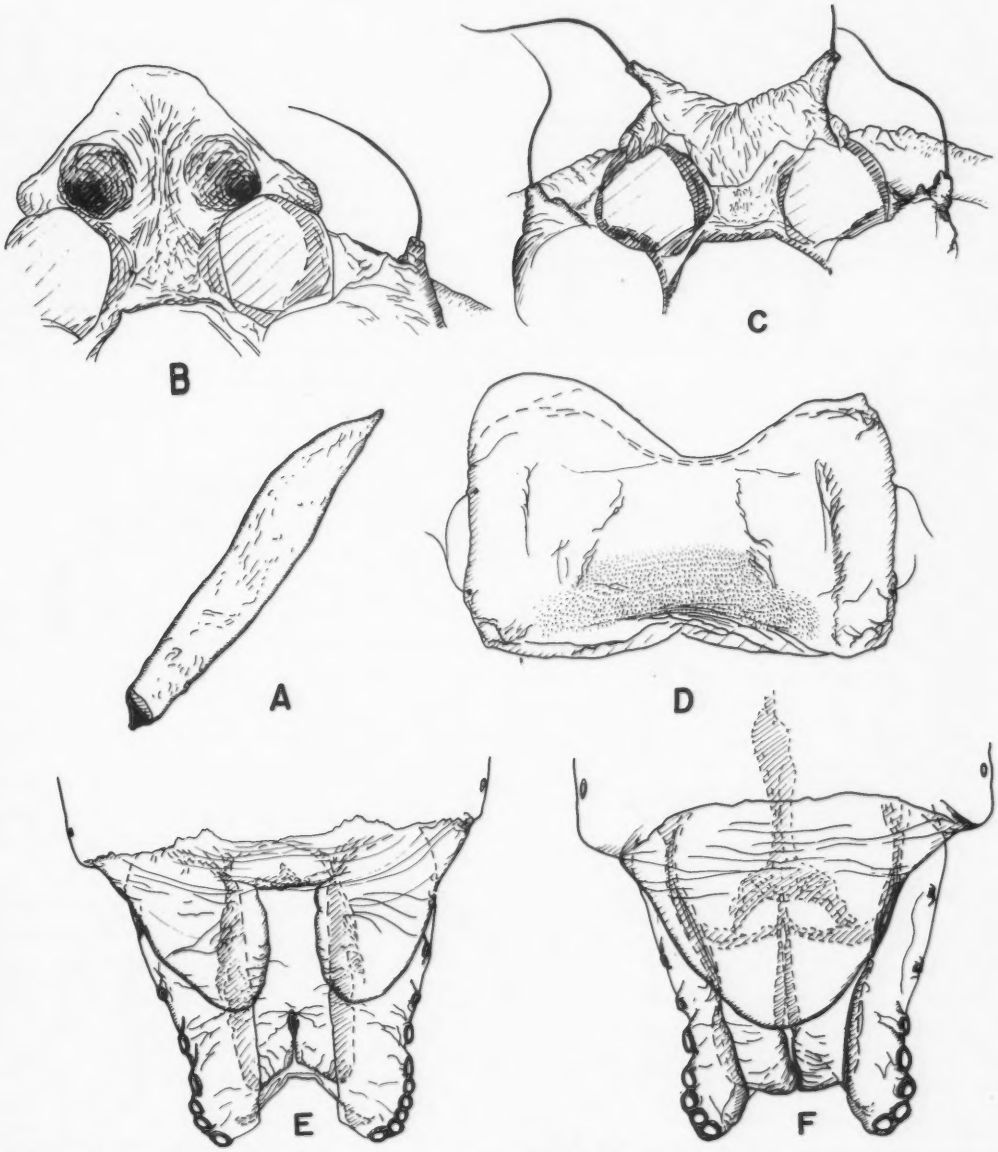

FIG. 6. *Protanypus hamiltoni* n.sp., pupa. A, thoracic horn; B, C, frontal plate, anterior plate, and anterior thoracic tubercle, different views; D, sternite I; E, F, caudal lobes of segment VIII and anal lobe of female (E) and male (F).

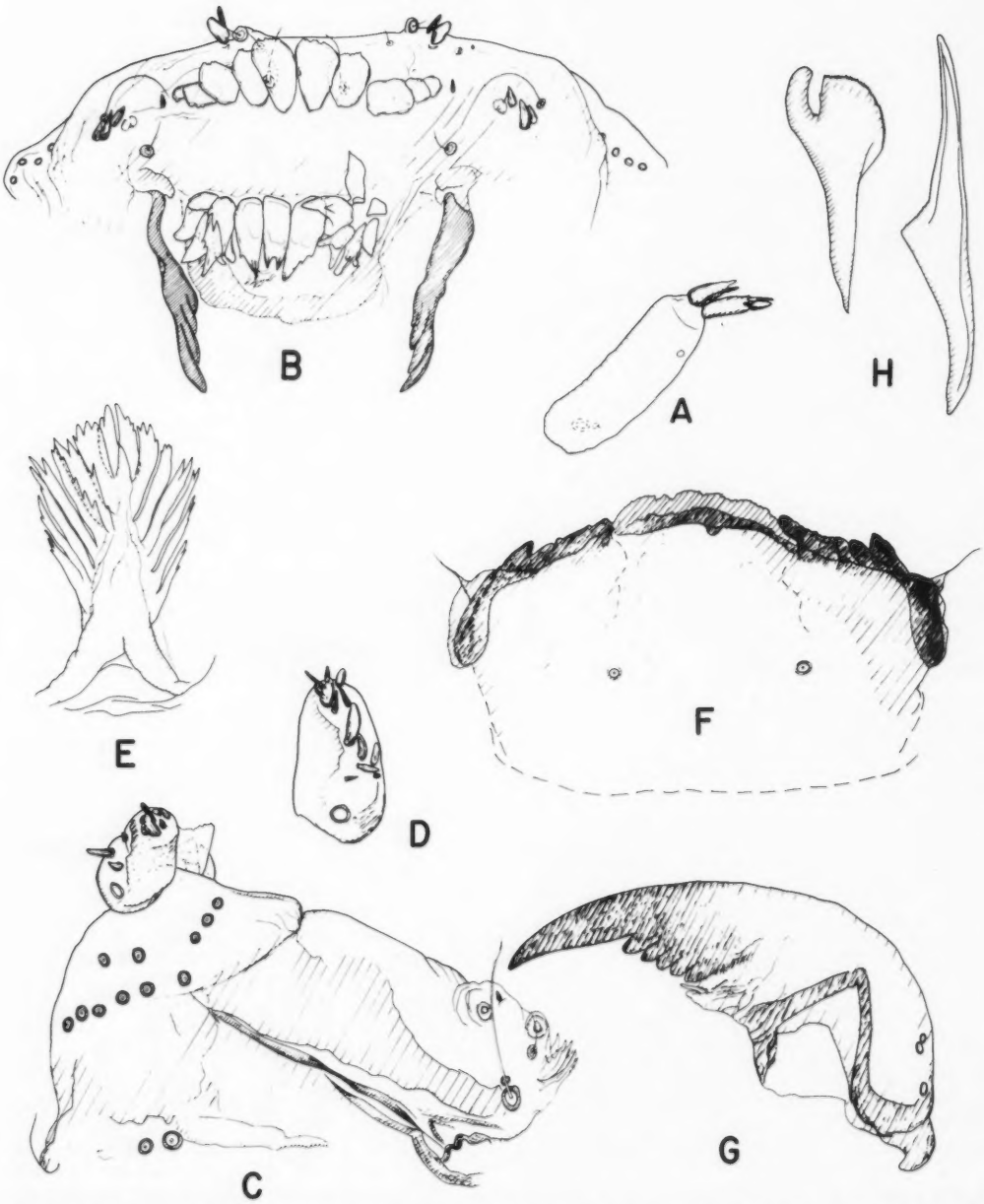

FIG. 7. *Protanypus hamiltoni* n.sp., larva. A, antenna; B, labrum and palatum; C, D, maxilla and maxillary palp, ventral (C) and dorsal view (D); E, medioventral appendix of prementum; F, mentum; G, mandible; H, claws of posterior parapods.

Remarks

The imago of this species as well as that of the preceding species closely resemble *P. forcipatus* (Egg.) from the Alps. *P. forcipatus*, however, according to Brundin (1952, p. 42, fig. 8), does not have microtrichiae on outer surface of the gonostylus, whereas both these species do (in proximal 0.4 in *P. ramosus* n.sp. and in proximal 0.8 in *P. hamiltoni* n.sp.) The leg ratio of the front leg is also mentioned to be only 0.63 in *P. forcipatus* (Brundin 1952, p. 43) while these species have leg ratios of 0.66–0.74. *Protanypus ramosus* n.sp. differs from *P. hamiltoni* n.sp. in the extent of microtrichiae of the gonostylus mentioned above, by having fewer setae on R and R_1, more numerous temporals (60–64 compared to 31–40), presence of distinct group of preoculars, metatarsus of front leg with 7–8 pseudospurs compared to only 2, lower BR_2, and intermedian endomere with a long apical tooth and 1–2 shorter lateral teeth compared to a short apical tooth and 1–2 short and 1 long simple to four-pronged lateral teeth in *P. hamiltoni*.

The pupae of both new species differ from those of *P. caudatus* Edw. and *P. forcipatus* (Brundin 1952, pp. 39, 43) by having the thoracic tubercles about half as large as the frontal tubercles, i.e. intermediate between *P. morio* Zett. and *P. forcipatus* plus *P. caudatus* (Brundin 1952, fig. 10–11), extensive shagreenation also on tergite II, and caudal lobe or lobes of segment VIII not sharply pointed. *Protanypus ramosus* differs from *P. hamiltoni* by having fine spinules on the thoracic horn and by having the posterior band of spinules on sternite I occupying only the posterior ⅛–⅓, while it occupies the posterior ½–⅔ in *P. hamiltoni*.

The larvae of the two new species are nearly identical to one another. They both differ from *Protanypus* sp. A and *Protanypus* sp. B described below by having 11–12 labral scales in fourth-instar larvae (17–22 in the other two species), and by having style of second antennal segment twice as long as the accessory style, while these are equal in *Protanypus* sp. A and the style only slightly longer in *Protanypus* sp. B. *Protanypus ramosus* differs from *P. hamiltoni* by having a lower AR (about 2.2 compared with 2.3–2.7), and by having the medioventral appendix of prementum about 2.4–2.5 times as wide at the widest as at the narrowest part (1.9–2.2 in *P. hamiltoni*). There probably are further differences in the maxillae, labrum, hypopharynx, and epipharyngeal area, but more material needs to be examined.

Protanypus ramosus and *P. hamiltoni* form a species pair possibly geographically separated from one another and forming a sister group of *P. forcipatus* and possibly *P. caudatus*.

Protanypus sp. A

Protanypus sp. A pro parte, Hamilton 1965; 52 (larva, in part).

This larva differs from that of the other three described species by having about 22 labral scales, an AR of only about 2.0 and style of second antennal segment as long as accessory style and only slightly longer than segment 3.

Fourth-Instar Larva (n = 1)

Length 12.02 mm. Head capsule length 0.92 mm.

Head — Antenna as in Fig. 8A. Lengths of antennal segments in microns: 84, 28, 4, 8. AR = 2.04. Basal antennal segment 28 μ wide, 3.00 times as long as wide, distance from base to annular organ 16 μ, to basal seta 24 μ, blade at apex 29 μ long, accessory blade 26 μ long. Apical style of second segment and accessory style both 5 μ long. Labrum and palatum as in Fig. 8B, labrum with 22 scales. Premandible 124 μ long. Maxilla as in Fig. 8C. Medioventral appendix of prementum (Fig. 8D) 128 μ long, 100 μ wide, minimum width at base 40 μ, maximum width/basal minimum width: 2.50. Supporting endoskeleton of appendix 140 μ wide. Mandible (Fig. 8F) 336 μ long, apical tooth 76 μ long. Postmentum 420 μ long.

Abdomen — Procercus 142 μ high, 44 μ wide, with 6 apical anal setae and 2 weak lateral setae. Anal tubules triangular, 300 μ long, 100 μ wide at base. Posterior parapods 638 μ long with two kinds of apical claws (Fig. 8G).

Material Examined

Two larvae, depths 1–4.5 m, Marion Lake, B.C. 1/4 and 24/6/64, A. L. Hamilton.

Remarks

This larva appears to be closely related to *P. morio* Zett. (Lenz 1925, p. 86, Thienemann 1944, p. 634, Greze 1953, p. 78, Pankratova 1970, p. 60). [The larva described by Zavrel (1926, p. 208) and referred to by Thienemann (1944) as *P. morio* more likely belong to *P. caudatus* [com-

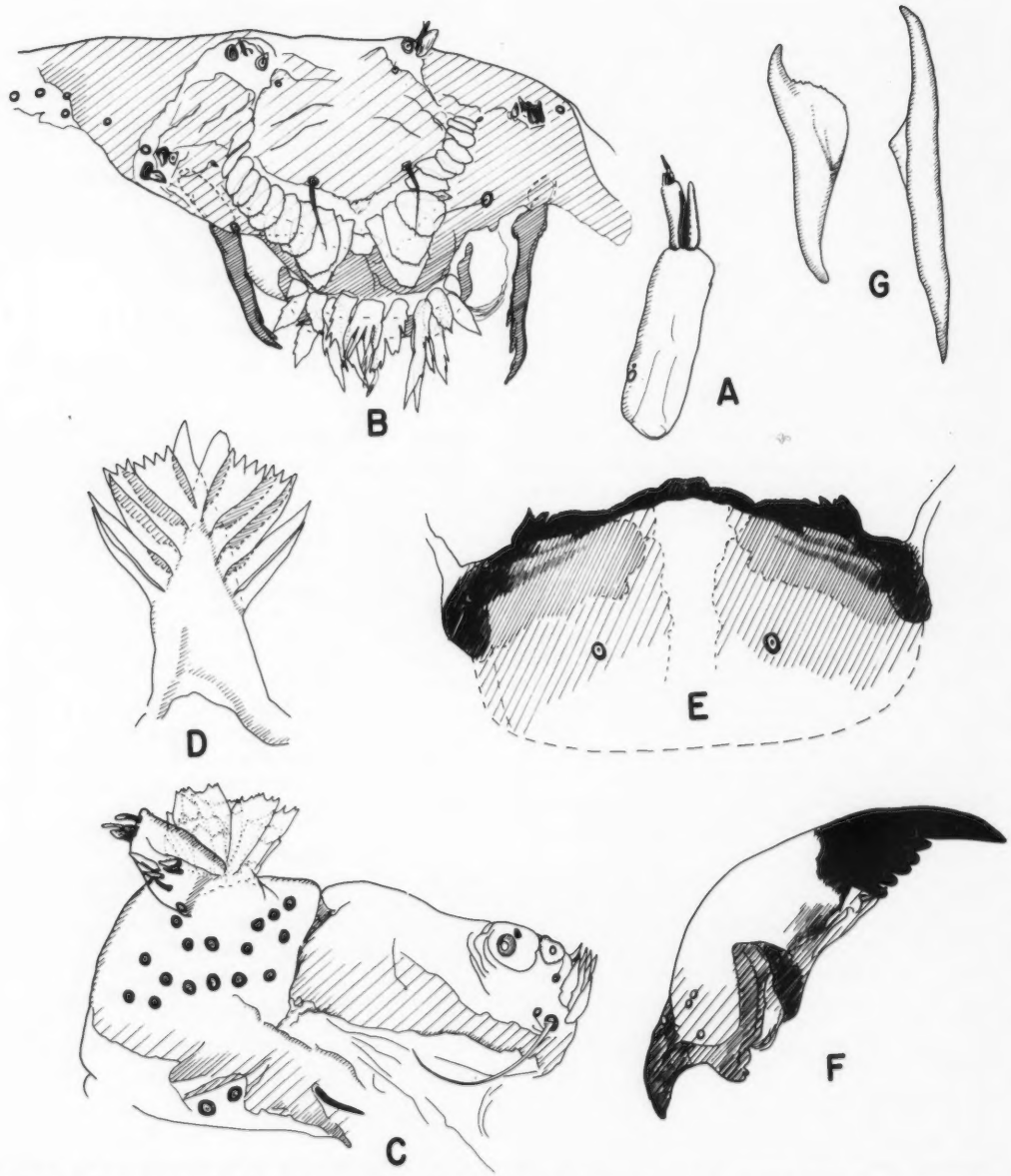

FIG. 8. *Protanypus* sp. A, larva. A, antenna; B, labrum and palatum; C, maxilla ventral view; D, medioventral appendix of prementum; E, mentum; F, mandible; G, claws of posterior parapods.

pare Zavřel (1926, fig. 3B) and Hirvenoja (1973, fig. 24,3).] However, *P. morio* is said to have only about 16–18 labral scales, an AR of about 3, antennal blade as long as the three last segments combined, and, if the tentative identification by Hirvenoja (1973, fig. 24, p. 43) is correct, slightly different medioventral appendix of prementum.

Protanypus sp. B

This larva differs from that of the other three described species by having 17 labral scales, an AR of about 2.9, and style of second antennal segment about 0.8 times as long as segments 3–4 combined and about 1.3 times as long as the accessory style.

FOURTH-INSTAR LARVA (N = 1)

Head capsule length 0.90 mm.

Head — Antenna as in Fig. 9A. Lengths of antennal segments in microns: 100, 25, 5, 7. AR = 2.86. Basal antennal segment 31 μ wide, 3.23 times as long as wide, distance from base to annular organ 8 μ, to basal seta 16 μ, to apical mark of seta 86 μ, blade at apex and accessory blade both 28 μ long. Apical style of second segment 10 μ long, accessory style 8 μ long. Labrum and palatum as in Fig. 9B, labrum with 17 scales. Premandible 110 μ long. Maxilla as in Fig. 9C. Medioventral appendix of prementum (Fig. 9D) 134 μ long, 84 μ wide, minimum width at base 42 μ, maximum width/basal minimum width: 2.00. Supporting endoskeleton of appendix 160 μ wide, and 2.29 times as wide as median tooth of mentum (Fig. 9E). Mandible (Fig. 9F) 286 μ long, apical tooth 86 μ long. Postmentum 384 μ long.

Abdomen – lost.

Material Examined

One larva, depth 17 m, George Lake, La Cloche Mountain Region, Ont., 23–30/9/68, R. J. Beamish.

Remarks

This larva appears to be closely related to *P. morio* Zett. and *Protanypus* sp. A (see p. 381). It differs from *P. morio* in having the antennal blade only slightly longer than second antennal segment and by having serrated median lamellae of medioventral appendix of prementum. *Protanypus* sp. A differs from this species in having 22 labral scales, an AR of about 2.04, apical style and accessory style of second antennal segment equal in length, and smooth median lamellae of medioventral appendix of prementum.

Key to Males of Nearctic and Palaearctic Species of *Protanypus*

1 Part of gonocoxite posterior to gonostylus about 0.5 times as long as gonostylus; gonostylus with microtrichiae on whole outer posterior surface, lateral endomeres smooth, without scales..... .....*P. morio* Zett. (Palaearctic)

Part of gonocoxite posterior to gonostylus 0.6 times as long to longer than gonostylus; gonostylus void of microtrichiae or with microtrichiae at proximal 0.4 or 0.8–0.9; lateral endomere with scales (not examined in *P. forcipatus*).....2

2 Part of gonocoxite posterior to gonostylus longer than gonostylus; outer surface of gonostylus without microtrichiae; LR = 0.76–0.81..... .....*P. caudatus* Edw. (Holarctic)[1]

Part of gonocoxite posterior to gonostylus about 0.6–0.8 times as long as gonostylus; outer surface with or without microtrichiae; LR = 0.61–0.74.....3

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[1]The males of the Nearctic species called *P. caudatus* by Oliver (1963, p. 177) have not been examined by the author. The females examined, however, have only about 7–10 sensillae chaeticae on the midleg and about 15–20 on the hind leg and the species thus is at least different from *P. ramosus* n.sp. There still is, however, some doubt whether the Nearctic imagines belong to *P. caudatus* or to a new undescribed Nearctic species.

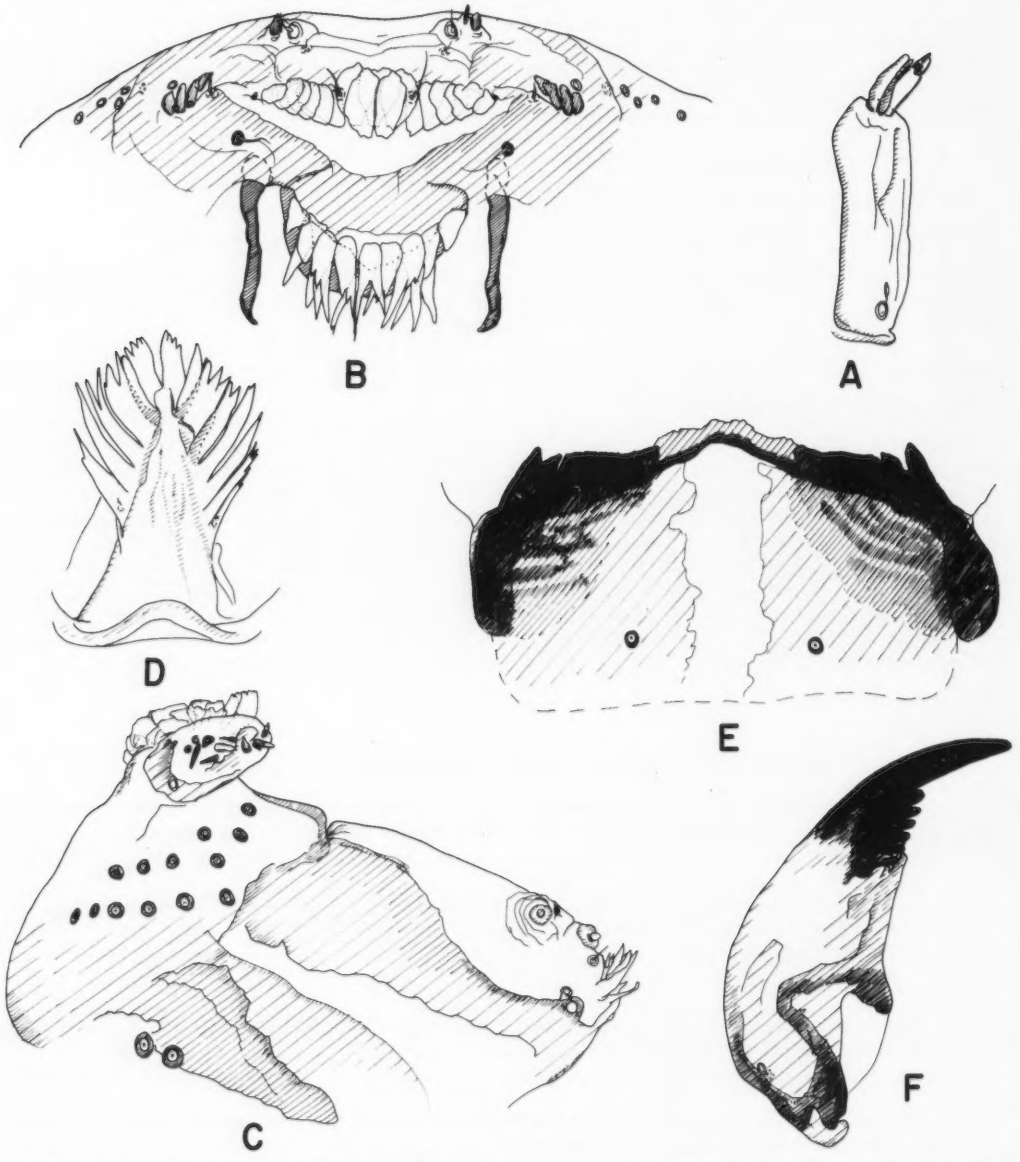

FIG. 9. *Protanypus* sp. B, larva. A, antenna; B, labrum and palatum; C, maxilla, ventral view; D, medioventral appendix of prementum; E, mentum; F, mandible.

3 Gonostylus void of microtrichiae on outer surface..... ..... *P. forcipatus* Egg. (Palaearctic)

Gonostylus with microtrichiae in basal 0.4 or basal 0.8–0.9..... 4

4 Front leg with 7–8 pseudospurs on ta_1, about 60–64 temporals including 2–3 preoculars, R_1 with about 9–12 setae, intermedian endomere with long apical tooth and 1–2 short and simple lateral teeth, gonostylus with microtrichiae in proximal 0.4..... .....*P. ramosus* n.sp. (Nearctic)

Front leg with 2 apical pseudospurs on ta_1, about 31–40 temporals, preoculars absent, R_1 with 20–26 setae, intermedian endomere with short apical tooth and 1–2 short and 1 long simple to 4-pronged lateral teeth, gonostylus with microtrichiae in proximal 0.8–0.9..... .....*P. hamiltoni* n.sp. (Nearctic)

Key to Pupae of Nearctic and Palaearctic Species of *Protanypus*

1 Anterior thoracic tubercle about as high and strong as frontal tubercle; shagreenation of tergites strong and extensive; thoracic horn 0.60–0.80 mm long, void of spinules; caudal lobe(s) of segment VIII broad and apically rounded.....*P. morio* Zett. (Palaearctic)

Anterior thoracic tubercle about 0.3–0.5 times as high as frontal tubercle; shagreenation of tergites weaker and less extensive; thoracic horn 0.30–0.70 mm long, with or without spinules; caudal lobe(s) of segment VIII more narrow, apically pointed, rounded or truncate.....2

2 Caudal lobes of segment VIII apically pointed, tergite II practically without shagreenation, tergite III with extensive shagreenation.....3

Caudal lobes of segment VIII apically truncate to rounded, tergite II with shagreenation nearly as extensive as on tergite III.....4

3 Thoracic horn short, at most 0.30 mm long, occasionally with a few distal spinules, but usually void of spinules..... .....*P. forcipatus* (Egg.) (Palaearctic)

Thoracic horn 0.45–0.60 mm long, with distal spinules..... .....*P. caudatus* Edw. (Holarctic)[2]

4 Sternite I with thin spinules up to 6 μ long, occupying posterior $\frac{1}{5}$–$\frac{1}{3}$; thoracic horn with weak, scattered spinules; caudal lobe of segment VIII usually apically truncate..... .....*P. ramosus* n.sp. (Nearctic)

Sternite I with spinules up to 12 μ long, occupying posterior $\frac{1}{2}$–$\frac{2}{3}$; thoracic horn without spinules; caudal lobe of segment VIII apically rounded.....*P. hamiltoni* n.sp. (Nearctic)

Key to Known Larvae of Nearctic and Palaearctic Species of *Protanypus*

1 16–22 labral scales, median lamellae of medioventral appendix of prementum smooth or serrated.. 2

10–12 labral scales, median lamellae of medioventral appendix of prementum serrated.....4

[2]See Footnote 1.

2 About 22 labral scales, AR about 2.2, blade at apex of basal antennal segment only slightly longer than second segment, style and accessory style of second antennal segment both only slightly longer than third segment, median lamellae of medioventral appendix of prementum smooth.....
.....*Protanypus* sp. A (Nearctic)

About 16–18 labral scales, AR about 2.9–3.0.....3

3 Blade at apex of basal antennal segment about as long as segments 2–4 combined, median lamellae of medioventral appendix of prementum smooth.....*P. morio* Zett. (Palaearctic)

Blade at apex of basal antennal segment only slightly longer than second segment, style of second antennal segment about 1.3 as long as accessory style and about 0.8 times as long as segments 3–4 combined, median lamellae of medioventral appendix of prementum serrated.....
.....*Protanypus* sp. B (Nearctic)

4 AR about 2.0, Holarctic species[3].....
.....*Protanypus* sp.prob. *caudatus* Edw. (Zavřel 1926, p. 208 as *Didiamesa* sp.).

AR about 2.2–2.7, style of second antennal segment about twice as long as accessory style and about as long as segments 3–4 combined, Nearctic species.....5

5 Medioventral appendix of prementum 2.4–2.5 times as wide as the minimum width at base, supporting endoskeleton of appendix 1.6–1.7 times as wide as median tooth of mentum, AR about 2.2
.....*P. ramosus* n.sp. (Nearctic)

Medioventral appendix of prementum 1.9–2.2 times as wide as the minimum width at base, supporting endoskeleton of appendix 1.3–1.6 times as wide as median tooth of mentum, AR = 2.3–2.7
.....*P. hamiltoni* n.sp. (Nearctic)

Distribution and Ecology

While Lenz (1925, p. 94) regarded the Boreoalpine *P. morio* as characteristic for mesotrophic lakes, Brundin (1949, p. 719; 1956, p. 203) showed that the species is most common in oligotrophic lakes. However, weak populations of *P. morio* may occur even in moderately eutrophic lakes, showing that the environment is not too extreme. *Protanypus morio* is bivoltine in Fennoscandia, but is a univoltine spring species in Lake Bodensee, the only certain record from the Alps (Reiss 1968, p. 228).

Protanypus caudatus is so far recorded only from strongly oligotrophic Fennoscandian lakes. However, of the several records of *Protanypus* larvae (see Pagast 1947, p. 561), the specimens from Lake Wigry and Lake Hańcza in northeastern Poland (Zavřel 1926, p. 208) appear to belong to *P. caudatus* according to the description (and not to *P. morio* as suggested by Brundin 1952, p. 43). The main basin of Lake Wigry is moderately oligotrophic, while Lake Hańcza is the most oligotrophic lake in the area. *Protanypus caudatus* is a bivoltine species (Brundin 1949 table 106, 1952 p. 41).

Protanypus forcipatus forms the Alpine sister species of the Boreal *P. caudatus* (Brundin 1952, p. 43). The species has been recorded only from oligotrophic lakes.

The two new Nearctic species are closely related to the species pair *caudatus–forcipatus*, and their ecology may be expected to be similar.

Protanypus ramosus was found at depths of 9–26 m in some of the most oligotrophic and unproductive lakes in the Experimental Lakes Area in Kenora, Ont. (*Protanypus* sp. in Hamilton 1971, p. 262). It is also present in Costello Lake, Algonquin Park, Ont. (*Protanypus* sp. in Miller 1941, p. 39), in South Indian Lake, Man. at 4 m depth, and at a depth of 43 m in Georgian Bay, Lake Huron (Brinkhurst et al. 1968, p. 19 as *Protanypus* cf. *forcipatus*). The larvae of *Protanypus* mentioned by Hiltunen (1969, p. 128) from Lake Superior probably also belong to *P. ramosus*. All these lakes are strongly oligotrophic and range from oligohumic to moderately polyhumic. The species is bivoltine with a strong

[3]See Footnote 1.

spring emergence in the first half of May and an apparently weaker fall emergence from the end of August to the beginning of October.

Protanypus hamiltoni is found in three deep and large oligotrophic lakes in British Columbia, namely Babine, Okanagan, and Kalamalka lakes, and at the shore of Great Slave Lake in the Northwest Territories. In Babine Lake the species was present in the littoral zone from where the only samples taken at the time were collected (Hamilton 1965, p. 55). In Okanagan Lake the species was found at depths of 13–117 m, in Kalamalka Lake at depths of 37–100 m (Sæther 1970, table 6 as *P.* cf. *morio*, Sæther and McLean 1972, table 3 as *P.* cf. *morio*). There may be several reasons why *P. hamiltoni* does not appear in the littoral zones of Okanagan and Kalamalka lakes, whereas it is present in the littoral zone of Babine Lake. One of these may be climatic, Babine Lake being considerably farther north; another may be the presence of pollution in the two other lakes which has affected the littoral zones to a considerable extent while the profundal zone is virtually unchanged. This species is also bivoltine with emergence in May and in September.

Protanypus sp. A. has so far been found only in Marion Lake, B.C. (Hamilton 1965, p. 52), a small and relatively shallow oligotrophic lake. Here it was collected at depths of 1 and 4.5 m.

Protanypus sp. B. has been found only in George Lake, Ont., an oligotrophic lake acidified by sulfur dioxide emitted by the smelters of Sudbury, Ont. (Beamish and Harvey 1972).

The four described species of *Protanypus* thus seem to have the same ecology as their European counterparts and can be regarded as important members of ultra-oligotrophic and moderately oligotrophic benthic communities in North America.

Acknowledgments

I am much indebted to Dr A. L. Hamilton, Department of the Environment, Fisheries and Marine Service, Freshwater Institute, Winnipeg, Man., for the types of *P. hamiltoni* and for the larval material of *P. ramosus* and *Protanypus* sp. A; to Dr D. W. Webb, Illinois Natural History Survey, Urbana, Ill., for the allotype and paratypes of *P. ramosus*; to Dr J. E. Sublette, Eastern New Mexico University, Portales, N.M., for a paratype of *P. hamiltoni*; to Dr R. J. Beamish, Department of the Environment, Fisheries and Marine Service, Pacific Biological Station, Nanaimo, B.C., for the larva of *Protanypus* sp. B; to Miss M. P. McLean for making most of the preparations, and to Mrs C. A. Galloway for the drawings.

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Midsummer Crustacean Zooplankton Communities in Acid-Stressed Lakes

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SPRULES, W. G. 1975. Midsummer crustacean zooplankton communities in acid-stressed lakes. J. Fish. Res. Board Can. 32: 389–395.

The distribution of limnetic crustacean zooplankton species and species associations in 47 industrially acidified lakes of the La Cloche Mountains, Ontario, are examined. pH, which ranged from 3.8 to 7.0, and to a lesser extent lake area and depth are the major determinants of the structure of these communities. *Mesocyclops edax*, *Cyclops bicuspidatus thomasi*, *Diaptomus minutus*, *Holopedium gibberum*, *Diaphanosoma leuchtenbergianum*, and *Bosmina* sp. were common species which occurred over the entire pH range. *Tropocyclops prasinus mexicanus*, *Epischura lacustris*, *Diaptomus oregonensis*, *Leptodora kindtii*, *Daphnia galeata mendotae*, *D. retrocurva*, *D. ambigua*, and *D. longiremis* plus some other rare species making a total of 64% of all species found never or rarely occurred below pH 5.0. *Polyphemus pediculus*, *Daphnia catawba*, and *D. pulicaria* were found primarily in lakes with low pH. An increase in the complexity of the zooplankton communities with increasing pH was evident. Above pH 5.0 the communities contained 9–16 species with three or four dominant species; in lakes with pH 5.0 or less the communities comprised one to seven species with only one or two dominants. Compared with relatively unspoiled lakes of similar morphometry in northwestern Ontario (ELA lakes) the La Cloche lakes have unusually simple zooplankton associations, particularly the very acidic lakes. Recurrent group analysis of the La Cloche and ELA lakes indicated that they were both characterized by a single major recurrent group of six species, five of which were common to the two areas. In the La Cloche lakes the group of six species occurred most frequently in lakes with pH above 5.0 although it maintained its integrity even down to pH 5.0. It is concluded that pH has a great effect on these zooplankton communities, primarily in lakes with pH below 5.0 where many species are completely eliminated and even tolerant species become progressively rarer until in some lakes a single species (*D. minutus*) remains.

SPRULES, W. G. 1975. Midsummer crustacean zooplankton communities in acid-stressed lakes. J. Fish. Res. Board Can. 32: 389–395.

L'auteur étudie la répartition des espèces de crustacés du zooplancton limnétique et les associations d'espèces dans 47 lacs des montagnes La Cloche, Ontario, rendus acides par l'industrie. La structure de ces communautés est en grande partie déterminée par le pH, qui varie entre 3.8 et 7.0, et, à un degré moindre, par la superficie et la profondeur du lac. *Mesocyclops edax*, *Cyclops bicuspidatus thomasi*, *Diaptomus minutus*, *Holopedium gibberum*, *Diaphanosoma leuchtenbergianum* et *Bosmina* sp. sont les espèces les plus communes qui se rencontrent à tous les pH. *Tropocyclops prasinus mexicanus*, *Epischura lacustris*, *Diaptomus oregonensis*, *Leptodora kindtii*, *Daphnia galeata mendotae*, *D. retrocurva*, *D. ambigua* et *D. longiremis*, de même que quelques autres espèces rares, représentant 64% de toutes les espèces trouvées, se rencontrent rarement ou jamais à un pH inférieur à 5.0. *Polyphemus pediculus*, *Daphnia catawba* et *D. pulicaria* se trouvent surtout dans les lacs à bas pH. Les communautés zooplantoniques deviennent plus complexes à mesure qu'augmente le pH. A un pH supérieur à 5.0, les communautés contiennent de 9 à 16 espèces, dont trois ou quatre sont dominantes; dans les lacs à pH de 5.0 ou moins, les communautés comprennent d'un à sept espèces, dont un ou deux seulement sont dominantes. Comparativement à des lacs de morphométrie semblable et relativement intouchés du nord-ouest de l'Ontario (lacs de la région des lacs expérimentaux), les lacs de La Cloche possèdent des associations zooplanctoniques exceptionnellement simples, surtout les lacs très acides. Une analyse des groupes récurrents des lacs de La Cloche et de ceux de la région des lacs expérimentaux révèle que, dans les deux régions, ces lacs sont caractérisés par un seul groupe récurrent de six espèces, dont cinq sont communes aux deux régions. Dans les lacs de La Cloche, le groupe de six espèces se rencontre le plus fréquemment dans des lacs dont le

Printed in Canada (J3533)
Imprimé au Canada (J3533)

pH est supérieur à 5.0, bien que le groupe demeure entier à un pH aussi bas que 5.0. L'auteur en conclut que le pH exerce sur ces communautés planctoniques une grande influence, surtout dans les lacs à pH inférieur à 5.0, d'où plusieurs espèces ont été complètement éliminées et où même les espèces tolérantes deviennent progressivement plus rares, au point que, dans certains cas, une seule espèce (*D. minutus*) demeure.

Received August 21, 1974
Accepted December 6, 1974

Reçu le 21 août 1974
Accepté le 6 décembre 1974

ACID fallout in rain and snow, apparently as a consequence of sulfur dioxide emission from Sudbury metal smelters, has led to rapidly increasing levels of acidity and associated reductions in the numbers of fish species in many lakes of the La Cloche Mountains, Ontario (Beamish and Harvey 1972). pH may have had a similar effect on the limnetic crustacean zooplankton communities in these same lakes (Sprules 1974). Stepwise linear multiple regression analysis of the number of zooplankton species on 28 physical and chemical variables indicated a strong positive association with pH which alone accounted for 53% of the variance in plankton species numbers; a positive association with lake length, interpreted as a measure of lake size, which accounted for an additional 18%, and a negative association with maximum depth which accounted for another 7%. Many of the zooplankton communities comprised fewer than four species, some only a single species, and the statistical results indicated that these simple associations occurred principally in acid lakes and to a lesser extent in small, deep lakes. Although characteristic of high altitude lakes and ponds (Pennak 1957; Anderson 1974), such simple zooplankton communities are unusual for Canadian Shield lakes, particularly those comparable in size and depth to the acid La Cloche lakes. The purpose of this paper is to extend the purely statistical analysis of zooplankton community structure by examining the distribution of individual species and species associations of the La Cloche lakes in relation particularly to pH and to a lesser extent to lake size and depth. These communities will also be compared to those of another group of Canadian Shield lakes which are not acid-stressed and for which detailed data exist (Patalas 1971).

Methods

Forty-seven lakes lying in a series of quartzite ridges of the La Cloche Mountains in and near Killarney Provincial Park, Ont., were surveyed during the summers of 1971–73 (Fig. 1). Detailed limnological data comprising 13 physical variables of lake size and depth, 15 chemical variables including total dissolved solids, hardness, alkalinity, pH, CO_2, O_2, sulphate, phosphate, and Secchi disc transparency, and total number of fish species were provided by H. H. Harvey. During the period of investigation a single field determination of pH was made for each lake from one water sample taken 0.5 m below surface at the center of the lake (see Harvey 1974 for further details of sampling techniques). Zooplankton samples were collected from July to early September with a conical tow net of 30 cm mouth diameter and 75-μ nylon mesh in 1972 and one of 25 cm mouth diameter and 110-μ mesh in 1973. There was no evidence that species numbers or composition was affected by net differences. Each lake was visited only once during the 2 yr and samples were taken during daylight hours from near bottom to surface in each major basin at the point of maximum depth. Sampling for zooplankton and water chemistry was always done in the same year, although not necessarily on the same day. At least $\frac{1}{15}$ of each zooplankton sample, or of pooled samples from multibasin lakes, was examined to identify and enumerate limnetic crustacean zooplankton species. Larger subsamples were examined for rare species. Taxonomic keys by Edmondson (1959), Brooks (1957), Brandlova et al. (1972), and Czaika and Robertson (1968) were used for identifications.

The recurrent group analysis (Fager 1957; Fager and McGowan 1963) is based on an index of affinity between species taken in pairs given as

$$I = [J/(N_A N_B)^{1/2}] - \tfrac{1}{2}(N_B)^{1/2}$$

where J is the number of lakes in which the two species cooccur, N_A is the number of lakes in which species A occurs, and N_B is the number of lakes in which species B occurs. Letters are assigned to species so that $N_A \leqslant N_B$. Only those pairs of species for which $I \geqslant 0.5$ are considered to show affinity. The largest possible group of species whose members show affinity with all other members of the group is formed, those species are eliminated, the next largest group of species is determined, and the procedure is continued until no further species remain or no further groups can be formed. The relationships amongst the groups are based on whether or not any members of one group show affinity with any members of another group.

Results and Discussion

In general the lakes are low in dissolved solids, calcium (2–23 ppm), and bicarbonate (1–20 ppm), and high in Secchi disc transparency

FIG. 1. Locations of the sampled lakes. Numbered lakes are: 1, Lesser Mountain; 2, Partridge; 3, Pellucid; 4, Agony; 5, Anguish; 6, Animus; 7, Logboom; 8, Lumsden V; 9, Lumsden IV; 10, Lumsden III; 11, Lumsden II.

(Table 1). Of particular note is that many of the lakes have low pH, one (Lesser Mountain) below 4.0 and many below 5.0. These lakes tend to lie in the southern and eastern portions of the La Cloche Mountains in the direction of the prevailing winds from Sudbury, whereas those with pH above 5.0 are located primarily in the northern and western portions away from the prevailing winds (Fig. 1). There is a strong negative correlation between lake pH and distance from Sudbury (Harvey 1974 and unpublished data).

The zooplankton species of these lakes are generally typical of Canadian Shield lakes. *Diaptomus minutus* occurs most frequently, inhabiting all but two lakes where it is replaced by *D. reighardi* (Table 2). *Holopedium gibberum* and *Bosmina* occur in about 75% of the lakes and *Mesocyclops edax*, *Cyclops bicuspidatus thomasi*, and *Diaphanosoma leuchtenbergianum* are less common, inhabiting about half the lakes. All of these species are dominant, in the sense of Patalas (1971), much more frequently than the remaining species. Daphnids occur less frequently and are rarely numerically dominant. *Daphnia* sp. which did not key out to a recognizable species, showed characteristics of both *D. rosea* and smaller species such as *D. parvula* or *D. ambigua*.

The most obvious trends in the distribution of individual species and species associations in the lakes are associated primarily with pH and to a much lesser extent with lake area and depth. This is particularly evident in Table 1 where the lakes have been arranged in order of decreasing pH. A discontinuity in species distribution occurs at a pH of 5.0–5.2 (Table 1, Fig. 2) and on this basis the species can be divided into three categories. The first, characterized by tolerant species distributed without regard to pH, includes *M. edax*, *C. b. thomasi*, *D. minutus*, *H. gibberum*, *Diaphanosoma leuchtenbergianum* and *Bosmina*, although some of these are rare at low pH. A second group including *Tropocyclops prasinus mexicanus*, *Epischura lacustris*, *Diaptomus oregonensis*, *Leptodora kindtii*, *Daphnia galeata mendotae*, *D. retrocurva*, *D. ambigua*, and *D. longiremis*, never occurs below pH 5.0. Some additional rare species are also included in this group so that of a total of 28 species found in the La Cloche lakes, 18 (64%) never or rarely occur below pH 5.0. A third group includes *Polyphemus pediculus*, *Daphnia catawba*, and the rarer *D. pulicaria* which occur primarily in lakes with pH below 6.0. The remaining species are too rare to be meaningfully included in these categories. These groups correspond comparatively well to those identified by Carter (1971) for ponds near Georgian Bay except that he found *Diaptomus minutus*, which was rare, only at higher pH's. *Diaptomus reighardi*, the common small calanoid copepod in Carter's ponds, was distributed without regard to pH, as is *D. minutus* in the La Cloche lakes.

There is a distinctive increase in the complexity of the zooplankton associations with increasing pH (Tables 1 and 3). Because many species do not survive below a pH of about 5.0 (Fig. 2), a rather abrupt change from simple to complex communities occurs at this point (Table 1). Most lakes with pH above 5.0 have from 9 to 16 zooplankton species with *D. minutus*, *C. b. thomasi*, and one or two of *Bosmina*, *H. gibberum*, and *Diaphanosoma leuchtenbergianum* dominant (Table 3). One exception to this is Little Sheguindah lake which has five dominant species but a pH of only 5.0. This may simply reflect sampling error but the lake is also interesting in that it is one of two which lie in a granite rather than quartzite basin, and contains *Diaptomus reighardi* and *M. edax* rather than *D. minutus* and *C. b. thomasi* as the dominant copepods. A. Y. Jackson is the other lake with a granite basin but it has a more typical community of 7 species with *Bosmina*, *H. gibberum*, *D. minutus*, and *C. b.*

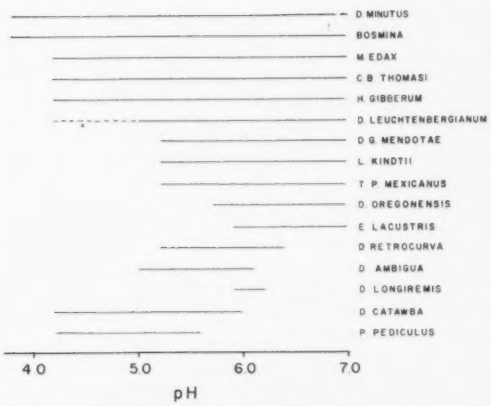

FIG. 2. Distribution of zooplankton species occurring in more than 10% of the lakes in relation to pH.

thomasi as dominants. The only other lake containing *D. reighardi* and *M. edax* as dominant copepods is Bodina which resembles Little Sheguindah insofar as it is small, shallow, and turbid. Additional data are required to determine whether these copepods are typical dominants of shallow, turbid lakes. The occurrence of only five species in Partridge and Patten lakes, even though each has a pH above 5.0, is probably due to the secondary effect of small surface area on the structure of the communities indicated by the regression analysis (Sprules 1974).

With the exception of Carlyle lake, which has nine species, lakes with pH 5.0 or less contain 1 to 7 species of midsummer zooplankton and two-thirds contain fewer than five. The majority of the lakes with pH 5.0 or less have either a single dominant species, *D. minutus*, or two dominant species including *D. minutus* plus one of *Bosmina*, *H. gibberum*, or *Diaphanosoma leuchtenbergianum* (Table 3). *Bosmina* is the single dominant species in Freeland lake, the shallowest lake in the survey with considerable rooted aquatic vegetation. Five of the lakes contained only a single species (*D. minutus*) when sampled, six others contained only two species, and four additional ones, including Killarney with a surface area of 357 ha and a maximum depth of 61 m, had only three. *Diaptomus minutus*, *Bosmina*, and *H. gibberum* occur most frequently and, on the average, most abundantly in lakes with pH 5.0 or less and hence are the most acid tolerant species. Norway, Animus, Anguish, Agony, and Pellucid, the lakes containing the single species *D. minutus*, have low pH (4.2–4.7) but are also relatively small and deep, notably Pellucid with a surface area of 13.8 ha and a maximum depth of 42.7 m. The secondary effects of depth and surface area on zooplankton community structure may partially explain why these lakes have only a single zooplankton species.

The unusual structure of many of the La Cloche zooplankton communities can best be appreciated by comparing them with limnetic crustacean zooplankton communities of 45 lakes, which are not acid-stressed, in the Experimental Lakes Area (ELA) of northwestern Ontario (Patalas 1971). These lakes are similar to the La Cloche lakes in that they lie on the Canadian Shield and are composed of relatively low-conductivity waters, but are different insofar as they lie in granite rather than quartzite basins, have higher pH (5.6–6.7), have lower concentrations of sulphates and some trace metals, and include a higher proportion of small lakes and shallow lakes (Armstrong and Schindler 1971; Patalas 1971). Despite these differences the comparison is informative and the following comments are made on the basis of all 47 La Cloche lakes and all 45 ELA lakes.

Patalas found a total of 26 limnetic species in the ELA lakes, 18 of which also occur in the La Cloche lakes which have a total of 28 species. Species occurring in 10% or more of the lakes differ in the presence of *Diaphanosoma brachyurum* and *Cyclops vernalis* in the ELA lakes only and of *Polyphemus pediculus* and *Daphnia ambigua* in the La Cloche lakes only. *Tropocyclops p. mexicanus* occurs in only 20% of the La Cloche lakes compared with 80% of the ELA lakes, probably reflecting its susceptibility to extremely low pH noted here and by Carter (1971). Its low frequency of occurrence could also be related to the fact that small lakes which are its preferred habitat in the ELA area are comparatively rare in the La Cloche area. *Polyphemus pediculus* is completely absent from the ELA lakes, whereas it occurs in 26% of the La Cloche lakes although it is never numerically abundant. Often a littoral species, it may be more limnetic in the acidic La Cloche lakes because of reduced fish species diversity (Harvey 1974) and hence reduced predation pressures. There is also the possibility that it experiences some competitive advantage due to the reduced abundance in the acid lakes of other predaceous zooplankters such as *Leptodora* and *Epischura*. The occurrence of *D. minutus* in 96% of the La Cloche lakes compared with only 73% of the ELA lakes reflects its tolerance of the acid conditions characteristic of so many La Cloche lakes.

The ELA zooplankton communities contain a

TABLE 1. Zooplankton communities and selected limnological characteristics of the La Cloch

	Surface area (*ha*)	Max depth (*m*)	TDS (*ppm*)	Secchi disc visibility (*m*)	pH	Total no. species	*Bosmina*[a] sp.	*Ceriodaphnia reticulata* (Jurine)	*Daphnia ambigua* Scourfield	*D. catawba* Coker	*D. galeata mendotae* Birge	*D. longiremis* Sars	*D. parvula* (?) Fordyce	*D. pulicaria* Forbes	*D. retrocurva* Forbes
Hele	68.2	41.2	19	6.0	7.0	12	.21				.04				
Low	28.6	28.4	37	7.3	7.0	10	.01				.02				
Ishmael	65.4	19.8	20	5.1	6.4	10	.11				+				.03
Bodina	30.3	6.7	23	2.4	6.3	9	.09	.06							.08
Quartzite	13.7	34.5	18	7.6	6.2	9	.10				.05	.07			
Charlton	218.9	18.0	28	5.1	6.2	13	.11					.05			.06
Gem	14.2	19.2	18	5.0	6.1	12	.18		.01		.07				
La Cloche	1088.3	30.5	28	2.5	6.0	15	.10			+	+	+			.03
Howry	101.9	27.4	19	6.0	6.0	9	.19				.09	+			
Patten	8.7	6.4	22	6.0	6.0	5	.01			.20					
Evangeline	384.9	19.2	29	3.4	5.9	13	.02				.03	.07			.02
Frood	191.8	18.0	26	6.3	5.9	16	.10	+[b]	+		+	.08			.04
Fish	90.7	8.5	17	5.7	5.7	9	.04		+						+
Kakakise	118.9	30.5	19	10.1	5.6	7	.06								
Logboom	6.0	6.0	22	5.5	5.3	10	.86	+	+						
Bell	281.3	26.8	22	6.9	5.2	11	.17		+						
Partridge	9.2	16.8	24	12.4	5.2	5									
Threenarrows	947.9	51.9	18	6.4	5.2	13	.09		.02	.02	.07				+
Johnnie	395.5	33.5	23	9.3	5.1	7	.05			+					
Carlyle	166.9	14.6	18	7.6	5.0	9	.20		+						
Little Sheguindah	5.0	2.7	17	2.4	5.0	5	.16								
A. Y. Jackson	6.1	9.8	20	6.9	5.0	7	.14		+						
Lumsden III	5.3	7.6	16	4.3	5.0	3									
Lumsden IV	5.2	4.9	17	4.3	5.0	2									
Lumsden V	3.4	10.1	17	4.4	4.9	2									
George	147.9	30.5	18	9.0	4.9	4	.14								
O.S.A.	291.4	39.7	24	13.2	4.8	3	.15								
Muriel	22.6	12.2	23	7.1	4.8	6	+							.02	
Lumsden II	17.7	30.0	16	2.4	4.8	4				+					
Norway	59.6	33.6	22	12.5	4.7	1									
Killarney	357.2	61.0	23	11.3	4.7	3	.11								
David	325.8	24.4	21	12.9	4.6	6	.06						.07	.03	
Animus	4.6	21.6	20	10.3	4.6	1									
Anguish	10.5	28.7	24	13.1	4.6	1									
Turbid	14.8	9.4	25	7.3	4.6	4	.02							.22	
Agony	12.1	33.5	28	12.3	4.5	1									
Ruth-Roy	46.1	18.0	23	12.8	4.5	3									
Little Mountain	20.0	25.0	27	14.8	4.4	2	.10								
Great Mountain	191.6	37.5	20	8.6	4.4	5	.07								
Lumsden I	21.5	22.9	13	8.0	4.4	6	.40			.03					
Clearsilver	23.9	13.7	24	10.8	4.3	2	+								
Terry	10.1	8.0	19	8.0	4.2	7	.03			+					
Pellucid	13.8	42.7	22	14.0	4.2	1									
Kirk	74.1	9.8	18	9.5	4.2	5	.05							.09	
Freeland	32.2	1.8	24	>1.8	4.2	5	.95								
Nellie	238.3	54.9	24	20.9	4.1	2									
Lesser Mountain	16.6	16.8	32	10.2	3.8	2	.23								

[a]Probably all *Bosmina longirostris* (O. F. Müller) (C. Goulden personal communication).
[b]Less than .01.

a Cloche lakes. Proportionate numerical composition of the plankton communities is presented.

D. retrocurva Forbes	*D. schodleri* Sars	*Daphnia* sp.	*Diaphanosoma leuchtenbergianum* Fischer	*Holopedium gibberum* Zaddach	*Latona setifera* (O. F. Müller)	*Leptodora kindtii* Focke	*Polyphemus pediculus* (L.)	*Diaptomus reighardi* Marsh	*D. minutus* Lilljeborg	*D. oregonensis* Lilljeborg	*Epischura lacustris* Forbes	*Limnocalanus macrurus* Sars	*Senecella calanoides* Juday	*Cyclops bicuspidatus thomasi* Forbes	*C. scutifer* Sars	*Mesocyclops edax* (Forbes)	*Macrocyclops albidus* (Jurine)	*Orthocyclops modestus* (Herrick)	*Tropocyclops prasinus mexicanus* Keifer
			.04	.02		+			.12	.04	+		+	.52		+			+
			+	+		+			.61	.05	+			.30		+			
.03			.02	+					.20	+	+			.59		.03			
.08			.06	+				.23			+					.16			.32
			.02	.02		+			.31		+			.42					
.06			.13	.04		+			.25	.10	+		+	.04		.19			.03
			.17	.02		+			.15	+	+			.32		.07			+
.03			.07	+		+			.05	.02	+	+		.16		.40			.17
			.08	.06					.27	.01				.28		+			
				.07					.74							.15			
.02			.02	.01		+			.10	.40	+		+	.09		.24			
.04			.11	.08	+	+			.35	.03				.08		.09		.03	.01
+			.25	+					.52	.07						.07			.06
		+		+			+		.35					.56		.02			
			+	+			+		.10					+		.01	+		
			.03	.08		+	+		.41					.16	.02	.04			+
		+		+			+		.96					.04					
+			.07	.08			+		.49					.09	+	.04			+
			.01	.16					.76					.01		+			
	+	+	.20	.06					.51					.02		+			
			.32	.19				.19								.14			
			.03	.38					.25					.20				+	
				+					.71									.29	
				+					.99										
									.96									.05	
				.26			+		.60										
				.04					.82										
				.17			+		.80							+			
				+					.92					.08					
									1.00										
							+		.89										
				.14			+		.70										
									1.00										
									1.00										
				.38					.38										
									1.00										
				.05			+		.95										
									.90										
				.03					.84					.02		.04			
				+			+		.56					+					
									.99										
			+	.22					.58					.14		.03			
									1.00										
				.15					.67							.03			
			+	+			+		.04										
							+		.99										
									.77										

TABLE 2. Frequency of occurrence of zooplankton species.

	Number of lakes			
	In which species occurs	%	In which species is dominant[a]	%
Diaptomus minutus	45	96	41	87
Holopedium gibberum	36	77	9	19
Bosmina sp.	35	74	16	34
Mesocyclops edax	23	49	6	13
Cyclops bicuspidatus thomasi	22	47	11	23
Diaphanosma leuchtenbergianum	21	45	6	13
Polyphemus pediculus	13	28	0	0
Daphnia galeata mendotae	10	21	0	0
Diaptomus oregonensis	10	21	1	2.1
Tropocyclops prasinus mexicanus	9	19	2	4.3
Leptodora kindtii	9	19	0	0
Epischura lacustris	9	19	0	0
Daphnia ambigua	8	17	0	0
D. retrocurva	8	17	0	0
D. catawba	7	15	1	2.1
D. longiremis	6	13	0	0
D. pulicaria	4	8.5	1	2.1
Orthocyclops modestus	4	8.5	1	2.1
Daphnia sp.	3	6.4	0	0
Ceriodaphnia reticulata	3	6.4	0	0
Senecella calanoides	3	6.4	0	0
Cyclops scutifer	2	4.3	0	0
Diaptomus reighardi	2	4.3	2	4.3
Daphnia parvula (?)	1	2.1	0	0
D. schodleri	1	2.1	0	0
Latona setifera	1	2.1	0	0
Limnocalanus macrurus	1	2.1	0	0
Macrocyclops albidus	1	2.1	0	0

[a]Greater than 10% of total sample.

TABLE 3. Composition of the zooplankton communities by dominant species.

No. of dominants[a]	Lakes		Median pH	Most frequent composition			
	No.	%		Calanoids	Cyclopoids	Cladocerans	Species
1	17	36	4.6	1	0	0	*D. captomus minutus*
2	15	32	5.0	1	0	1	*D. minutus* + *Bosmina*, *Holopedium*, or *Diaphanosoma*
3	9	19	6.0	1	1	1	*D. minutus* + *Cyclops bicuspidatus thomasi* + *Bosmina*, or *Holopedium*
4	5	11	6.0	1	1	2	*D. minutus* + *C. b. thomasi* + *Bosmina* + *Holopedium* or *Diaphanosoma*
5	1	2	5.0	1	1	3	*Diaptomus reighardi* + *Mesocyclops* + *Bosmina* + *Holopedium* + *Diaphanosoma*

[a]Greater than 10% of all individuals.

mean of 8.5 species with only two lakes (4.4%) having four species and the remainder having at least five. In contrast the La Cloche communities contain a mean of 6.3 species with 14 (30%) lakes having fewer than four species. Only three (7%) ELA lakes had a single dominant species and three dominants was the most common situation, whereas 17 (36%) La Cloche lakes had one dominant species, the most common situation. Thus the ELA and La Cloche zooplankton communities are comparable in total number of species and, with the odd exception, in frequently occurring species. However, they differ considerably in numbers of species, the major difference being the comparative simplicity of many of the La Cloche communities in lakes with pH below 5.0. It is noteworthy that small deep lakes contained the simplest ELA communities and in the La Cloche area, next to low pH, the lake characteristics most frequently associated with simple communities were smallness and deepness.

Recurrent group analysis (Fager 1957; Fager and McGowan 1963) of the zooplankton communities in the two groups of lakes show interesting similarities (Fig. 3). These associations represent groups of species which occur in one another's presence in a relatively high proportion of the lakes. The most interesting point is that despite the differences in locality and chemistry, the ELA and La Cloche lakes are each characterized by a single major recurrent group comprising six species, five of which are common to the two areas. This similarity of characteristic species in the two areas supports Patalas' (1971) hypothesis of faunistic similarity for the regions. Although the species of the major recurrent group in the La Cloche lakes co-occur to some extent below pH 5.0, their more frequent co-occurrence in lakes with higher pH (Table 1) probably made the greater contribution to the formation of the recurrent group. Hence this association of species, which may be typical of Canadian Shield lakes of northern and central Ontario, occurs primarily in the less acidic La Cloche lakes but it is interesting that it maintains its integrity even in lakes with pH as low as 5.0. *Tropocyclops p. mexicanus*, a member of the ELA major recurrent group, does not appear in the major La Cloche recurrent group probably because of its complete absence from lakes with pH below 5.2 and its low frequency in lakes with higher pH. The six species in the major group of the La Cloche lakes are those which appear to be tolerant of the greatest range of acidity (Fig. 2). A second fairly large recurrent group comprising four species restricted to lakes with pH above 5.2 appears in the La Cloche lakes (Fig. 3). In fact, all remaining La Cloche species included in the analysis are restricted to higher pH's, although their co-occurrences are such that they form three groups with the interrelationships depicted in Fig. 3.

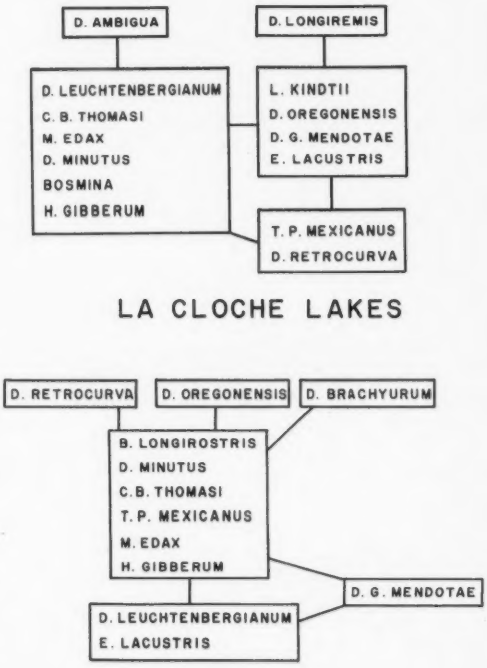

FIG. 3. Recurrent groups and their interrelationship for the La Cloche and ELA lakes. A line is drawn between groups when species in one group show affinities with some species in the other group.

Conclusions

The evidence presented suggests that pH is the factor which has the major effect on the structure of the La Cloche zooplankton communities. This effect is greatest below pH 5.0 where many species are completely eliminated and even tolerant ones become progressively rarer until a single species remains. Above this, pH has very little effect on the tolerant species and only a slight effect on the total number of species (Table 1). In fact, La Cloche lakes with pH above 5.0 contain an association of species very similar to the group of species characteristic of the relatively unspoiled ELA lakes of northwestern Ontario. In contrast,

La Cloche lakes with pH below 5.0 contain zooplankton communities of unusually low species numbers relative to the ELA lakes. *Diaptomus minutus*, the most acid-tolerant species in the La Cloche lakes, progressively increases in relative abundance as pH decreases and other species disappear until in five very acidic lakes it is the only species present. Carter (1971) and Salazkin (1971) also found that zooplankton communities became progressively less diverse as pH decreased.

The mode of action of pH, or a related variable, is unknown, as are the implications to whole-lake ecology of such simple zooplankton associations. Single species communities are characteristic of alpine (Anderson 1974) and arctic (Rigler et al. 1974) lakes but are very unusual for temperate lakes of the size and complexity of the La Cloche lakes. A reduced size range of primary consumers may reduce the feeding efficiency of secondary consumers (Kerr 1971) and hence affect community structure at that level. Alternatively, reduced numbers of zooplankton species imply that a smaller range of particle sizes will be ingested (Brooks and Dodson 1965) with possible effects on the phytoplankton (Hrbacek et al. 1961) and bacterial communities. There is some suggestion here and in Sprules (1974) that, aside from direct effects on the plankton, pH changes, by virtue of the effects on fish, would disrupt normal competitive interactions leading to accelerated changes in zooplankton communities. Until more is known about the effects of perturbations on aquatic ecosystems, unnatural inputs which precipitate changes of the sort documented here should be avoided.

Acknowledgments

I am indebted to Dr H. H. Harvey who kindly provided much of his raw limnological data, and to Mr H. Wener who assisted with the identification of the species. This research was supported by grants from the National Research Council of Canada, The Department of the Environment, and Erindale College, all of whose assistance is gratefully acknowledged.

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Embryonic Development, Early Growth, and Meristic Variation in Rainbow Trout (*Salmo gairdneri*) Exposed to Combinations of Light Intensity and Temperature[1]

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KWAIN, W. 1975. Embryonic development, early growth, and meristic variation in rainbow trout (*Salmo gairdneri*) exposed to combinations of light intensity and temperature. J. Fish. Res. Board Can. 32: 397–402.

Lowest mortality rates of rainbow trout (*Salmo gairdneri*) embryos were obtained at temperatures of 7 and 10 C and light intensities of 0.2 and 20 lx. Temperatures of 3 and 15 C and an intensity of 400 lx were near the thresholds for development. Eggs exposed to 0.2 lx required 111 days to reach 50% hatch at 3 C, but 26 days at 15 C; those exposed to 20 lx, 97 days at 3 C and 25 days at 15 C.

Growth rates of rainbow trout 145 days after hatch were significantly different ($P < 0.05$) at 10 and 3 C, and 20, 2, and 0.2 lx. The fastest growth occurred at 10 C and 2 lx, and the lowest growth at 3 C and 0.2 lx. For increment of body weight it was about 23.8%/day of initial weight and 6.6% at 3 C. At light intensities of 20, 2, and 0.2 lx, the rate per day was 24.7, 17.2, and 11.2%, respectively. However, increases in length occurred at a much reduced rate.

Variations in numbers of vertebrae, gill rakers, and fin rays were positively correlated with the embryonic development rate. Longer incubation periods were usually associated with more meristic elements, regardless of the environmental factors involved.

KWAIN, W. 1975. Embryonic development, early growth, and meristic variation in rainbow trout (*Salmo gairdneri*) exposed to combinations of light intensity and temperature. J. Fish. Res. Board Can. 32: 397–402.

Les plus faibles mortalités d'embryons de truite arc-en-ciel (*Salmo gairdneri*) se produisent à des températures de 7 et 10 C, et à des intensités lumineuses de 0.2 et 20 lx. Des températures de 3 et 15 C, et une intensité de 400 lx se rapprochent des seuils de développement. Les oeufs exposés à 0.2 lx requièrent 111 jours pour une éclosion de 50% à 3 C, mais 26 jours à 15 C; les oeufs exposés à 20 lx requièrent 97 jours à 3 C et 25 jours à 15 C.

Le rythme de croissance des truites arc-en-ciel, 145 jours après éclosion, diffère de façon significative ($P < 0.05$) à 10 et 3 C, et à 20, 2 et 0.2 lx. La croissance la plus rapide se produit à 10 C et 2 lx, et la plus lente à 3 C et 0.2 lx. En termes d'augmentation de poids corporel, elle est d'environ 23.8% par jour du poids initial et 6.6% à 3 C. A des intensités lumineuses de 20, 2 et 0.2 lx, les taux quotidiens sont de 24.7%, 17.2% et 11.2% respectivement. Cependant, la longueur augmente à un rythme considérablement diminué.

Les variations du nombre de vertèbres, de branchicténies et de rayons anaux sont en corrélation positive avec le rythme de développement embryonnaire. Des périodes d'incubation plus longues sont ordinairement associées à un plus grand nombre d'éléments numériques, quels que soient les facteurs ambiants en jeu.

Received August 26, 1974
Accepted November 29, 1974

Reçu le 26 août 1974
Accepté le 29 novembre 1974

THIS is a report on effects of combinations of temperature and light intensity on early development of rainbow trout (*Salmo gairdneri*): of five temperatures (3–15 C) and four intensities (0.2–400 lx) on embryonic survival and development rate; of two temperatures (3 and 10 C) and three intensities (0.2, 2, 20 lx) on growth to 145 days;

[1]Contribution No. 74-17, Ontario Ministry of Natural Resources, Fish and Wildlife Research Branch, Maple, Ont.

Printed in Canada (J3540)
Imprimé au Canada (J3540)

and of two temperatures (7 and 15 C) and four intensities (0.2–400 lx) on variations in several meristic characters. Much is known on effects of temperature on early development of fish, and some works on salmonids have suggested that light intensity may affect rate of embryonic development (e.g. Smith 1916) and meristic variation (Lindsey 1958). Temperature has been shown repeatedly to be one of the most influential environmental factors affecting the growth of fishes (Baldwin 1957; Brett et al. 1969; and many others), but less is known on effect of light intensities on growth. This is the first report on effects of combinations of the two variables on early development and growth in a salmonid.

Materials and methods — Newly fertilized eggs, from several females and males of hatchery origin, were obtained from the Ontario Provincial trout hatchery at Normandale. Eggs were incubated in an apparatus described by MacCrimmon and Kwain (1969). Two identical troughs, each with four compartments, were used. The light sources were four 500-W incandescent bulbs (3200 K). Light intensity in each compartment was regulated by neutral filters (exposed X-ray plates). The photoperiods were kept constant at 16 h light and 8 h dark.

Water temperatures were recorded continuously with a Dickson minicoder at a variation of ±.5 C. Light intensities were measured at the water surface with a low light level photometer (International Light Inc.). Eggs were submerged 15 cm below the water surface. Creek water was used without filtration: total alkalinity as $CaCO_3$ was 20 mg/liter; free carbon dioxide, 6 mg/liter; dissolved oxygen, 10 mg/liter; sodium, 1.0 mg/liter. Total phosphate, nitrogen–nitrate, and potassium were not detectable.

Each compartment contained about 2000 eggs, taken randomly from a stock of 20,000 newly fertilized eggs. Dead eggs were counted and removed daily. To inspect and remove eggs incubating at 0.2 lx a flashlight with a red lens was used. A batch of eggs was considered hatched when 50% of the surviving embryos had freed themselves completely from the chorion.

Initial lengths and weights of fry were taken as those at 15 days after hatching. Fifty fry were retained in each of the compartments in which they had hatched. Food was provided ad lib. Length and weight measurements were recorded on 20 fry in each compartment at monthly intervals during 145 days. Daily growth rate was expressed as a percentage of the initial measurement. Both total length and weight were compared. The mortality rate in each compartment was negligible.

A sample of 50 fry from each of the experimental conditions was held for 7 mo in a trough for meristic study. Water temperature fluctuated with stream water source. Light intensity was adjusted to provide about 50–100 lx using natural window light and fluorescent tubes. The fry (about 60–80 cm) were killed and preserved in 70% alcohol until X-rayed for meristic counts (Table 1) as described by MacCrimmon and Kwain (1969). Mortality of experimental fish was negligible.

The experimental results were subjected to analysis of variance. Multiple range test (Steele and Torrie 1960) was used to test for significance between means at $P = 0.05$, the percent mortality having been treated with arc-sine transformation.

Embryonic development — The lowest mortality ($P < 0.05$) occurred at temperatures of 7 and 10 C and light intensities of 0.2 and 20 lx (Fig. 1). At the highest light intensity, 400 lx, and at the extreme water temperatures (3 and 15 C), regardless of other factors, embryos were near the thresholds of development. Multiple range comparison among means indicated that these two factors were additive. For example, 100% mortality of embryos occurred at 3 C and 400 lx.

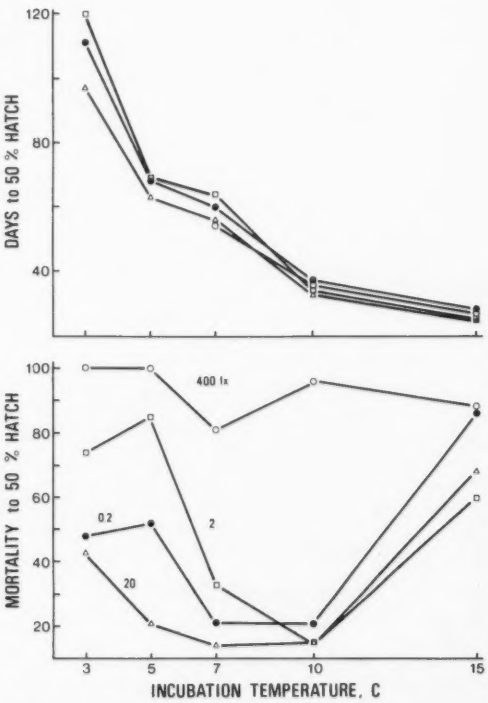

FIG. 1. Mortalities (%) and incubation periods of rainbow trout (*Salmo gairdneri*) eggs exposed to combinations of five temperatures and four light intensities.

TABLE 1. Mean counts for various meristic characters in rainbow trout (*Salmo gairdneri*) fry from eggs incubated at two temperatures (7, 15 C) and four light intensities (0.2, 2, 20, 400 lx).

				Vertebrae				
Conditions[a]	15,400	15,0.2	15,2	15,20	7,20	7,2	7,400	7,0.2
mean (0.0677)[b]	61.45	61.60	61.80	62.05	62.35	62.35	62.45	62.95
				Gill rakers				
Conditions[a]	7,400	7,0.2	15,2	7,20	7,2	15,400	15,20	15,0.2
mean (0.0742)[b]	18.10	18.10	19.30	19.40	19.50	19.50	19.70	19.70
				Dorsal fin rays				
Conditions[a]	15,400	15,20	15,2	15,0.2	7,20	7,2	7,400	7,0.2
mean (0.0182)[b]	13.00	13.00	13.00	13.00	13.00	13.00	13.20	13.60
				Pectoral fin rays				
Conditions[a]	7,0.2	7,400	15,0.2	7,20	7,2	15,400	15,2	15,20
mean (0.0221)[b]	13.65	13.90	13.90	13.95	13.95	14.00	14.00	14.10
				Pelvic fin rays				
Conditions[a]	15,400	15,0.2	15,2	15,20	7,400	7,20	7,2	7,0.2
mean (0.0202)[b]	9.15	9.60	9.85	10.00	10.00	10.00	10.00	10.00
				Anal fin rays				
Conditions[a]	15,400	15,0.2	15,20	7,2	7,20	15,2	7,400	7,0.2
mean (0.0396)[b]	11.15	11.30	11.85	12.00	12.10	12.15	12.20	12.50

[a]Temperature first and light intensity second.

[b]Standard error. Any two means not underscored by the same line differ at $P < 0.05$ (Student-Newman-Keuls' multiple range test; N = 160).

At 3 and 15 C, time to 50% hatch averaged 111 days and 26 days, respectively, indicating a fourfold increase in development rate (Fig. 1). However, the effect of light on development was less pronounced. A 100-fold increase in light intensity accelerated development by only 2 days at the highest temperature and by 14 days at the lowest. Analysis showed no significant difference among light intensities, but significant at various temperatures ($P < 0.05$). Multiple range test identified that development was higher at 5 and 7 C than at 3 C, and at 10 and 15 C than at lower temperatures.

Early growth — Growth rate of rainbow trout after feeding varied significantly at test temperatures and light intensities (Fig. 2a, b). Percent increase per day of initial measurements (15 days after hatch) between total length and weight was different, i.e. about 10 times greater in weight than in length. Percent increase of initial weight and length (in parentheses) per day at two temperatures and three light intensities were:

	20 lx	2 lx	0.2 lx
10 C	25.6 (1.9)	27.0 (2.1)	18.8 (1.5)
3 C	8.5 (1.3)	7.5 (1.4)	3.7 (1.0)

Fish held at 3 C and 0.2 lx were small in size and less in weight. Growth tended to be greater at 10 C and 2 lx, but no significant differences were found between 20 and 2 lx ($P > 0.05$).

Meristic variations — More meristic numbers were found at 3 C and 0.2 lx than at 10 C and at 400, 20 and 2 lx, except for gill rakers and pectoral fins (Table 1). The significance of the interaction of light and temperature indicates that these environmental factors are not operating independently of one another.

The mean vertebrae counts at 7 C were significantly higher than those at 15 C. On the other hand, the effect of light intensity on the total vertebral counts was not significant and showed no definite pattern. Concerning the incidence of fused vertebrae, they increased with increases in water temperature. In embryos incubated at 15 C, 16.18% of the fish were found to have fused vertebrae, but at 7 C, only 0.05%. The number of fused vertebrae ranged as high as five per fish at 15 C whereas at 7 C, the number rarely exceeded one per fish.

The mean counts of all fin rays increased significantly with decreases in water temperature, except for the pectoral fin. The effect of light on mean counts of all other fin rays was significant, but no clear linear trend was indicated. Mean

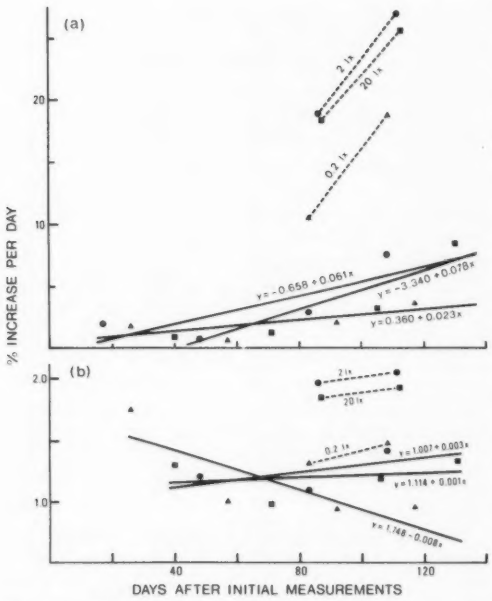

FIG. 2. Percentage daily increases in weight (*upper panel*: initial value 0.113 ± 0.018 g) and length (*lower*: initial value 2.04 ± 0.07 cm) for rainbow trout fry reared from 15 to 145 days after hatching at 3 C (*solid lines*) and 10 C (*broken lines*) and three light intensities (50 fry per test).

gill rakers and pectoral fin ray counts were significant with the higher mean counts occurring at 15 C. Again, the effect of light on mean counts of gill rakers and pectoral fin rays was significant, but no trend was indicated.

Discussion — Optimum condition for embryonic development of rainbow trout was between 7 and 10 C and at 0.2 and 20 lx. Any deviation from this condition will result in high mortality, either by temperature or light intensity. A similar optimum thermal condition was reported by Embody (1934), but no light intensity was given. Rainbow trout embryos were found to be tolerant of temperatures between 3 and 15 C and light intensities less than 400 lx. The thermal range agrees with results of Garside (1966), who reported a developmental range of 2.5–17.5 C for the same species. MacCrimmon and Kwain (1969) also found that rainbow trout held at 200 lx in a constant temperature of 9.2 C had 65% mortality. The present study demonstrated that effect of light intensity and temperature on embryonic development is additive, also light intensity and temperature can act singly and independently of the level of other factors.

The approximate hatching time of rainbow trout can be predicted from the relationship of development rate and temperature (Fig. 3), if all other environmental factors are favorable. However, in the present study embryonic development was not observed over the whole range of developmental temperatures. Rather, only the central linear segment of the curve was used in the study. In the interest of practicality, the developmental zero and upper thermal threshold were not considered. Generalizations concerning development rate were discussed by Kinne and Kinne (1962), and again by Garside (1966). They stated that development rate is not a linear function of temperature throughout the range of developmental temperature. However, the present study tended to ignore the inflections of such a curve and to consider only the central, linear segment.

Differences in growth following hatch were observed in samples of rainbow trout held at two temperatures (10 and 3 C) and three light intensities (20, 2, and 0.2 lx). The higher temperature always coincided with the higher growth rate (Fig. 2). For either temperature, the effect of three light intensities (20, 2, and 0.2 lx) on growth rate was significant. At low light intensity, fish grew slower than those at higher light intensities. This agrees with the findings of Bilton (1972) in studies on juvenile sockeye salmon, *Oncorhynchus nerka*; when reared in continuous darkness, mortality was very high, and the survivors were small. Similarly, rainbow trout reared at the lowest light intensity (0.2 lx) gain less length and weight than those at the higher light intensities (20 and 2 lx), regardless of the water temperature. However, no differences were de-

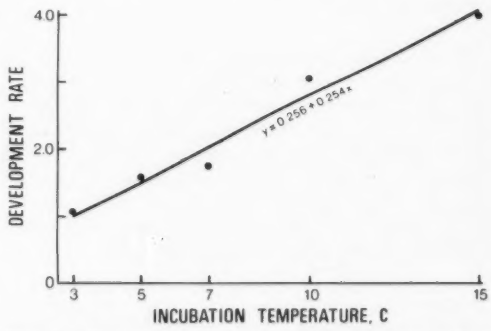

FIG. 3. Development rate of rainbow trout at five incubation temperatures at a light intensity of 20 lx.

tected in fish reared at 2 and 20 lx. The reasons for the slow growth at the lowest light intensity are not known but might be associated with their inability to see clearly. Fish were observed at the time of feeding by the use of a covered flashlight. Those reared at 0.2 lx were apparently sluggish and not actively looking for food, while those at higher light intensities actively sought out food.

This study shows that the number of serial elements is determined by developmental rate; longer developmental periods are usually associated with higher counts in meristic structure. Temperature is the main factor controlling the rate of development but light exerts some affect at a lesser degree.

An increase in vertebral number with increased time of development was obtained whether rates were governed by temperature or light. Considering temperature only, Hubbs (1926) stated that warmwater fishes generally have fewer vertebrae than do individuals of the same species inhabiting waters of lower temperature. In addition, Tåning (1944), working with sea trout *Salmo trutta*, and Orska (1962), working with rainbow trout, showed that temperature affects vertebral numbers during incubation, especially at the preeyed stage. The present findings are in agreement with the above mentioned earlier results. With respect to the effect of light intensity on the vertebral number of rainbow trout, MacCrimmon and Kwain (1969) found a positive relationship between vertebral number and light intensity when the temperature was held constant at 9.2 C. The present study showed that the mean vertebral counts at experimental conditions were significant but nonlinear, indicating that temperature and light are not acting independently even though temperature is the major factor involved. However, the percentage frequency of individuals with fused vertebrae increased with increase in temperature, which agrees with results reported by Canagaratnam (1959). Apparently, increases in the frequency of abnormal vertebrae coincided with accelerated developmental rate.

The mean number of fin rays was greater at lower temperatures with lower developmental rate, except for the pectoral fin. Barlow (1958) found the same result in Gobiid fish (*Gillichthys mirabillis*), i.e. the pectoral fins did not correlate to other fins. The effect of light on the mean number of fin rays was somewhat more complex. The highest number of fin rays occurred at 20 and 2 lx and the lowest at 400 and 0.2 lx, except for the dorsal fin where the relationship was reversed. The light conditions favoring high mean counts of fin rays could be interpreted as being most suitable for development, whereas conditions outside these levels would be less favorable for development.

Finally, some comment is warranted concerning the relation between the number of meristic characters and the rate of development. Hubbs (1926) and Gabriel (1944) hypothesized that the frequency of meristic characters is controlled through two different aspects of developmental physiology, growth, and differentiation. In this scheme, low temperatures (low developmental rate) retard growth more than differentiation. When the growth is slowed, the presumptive tissues presumably develop into a greater number of elements before the segments are fixed; conversely, rapid growth does not permit the differentiation of as many elements. The results in this paper seem to support this hypothesis reasonably well.

Acknowledgments — I am deeply indebted to Dr E. T. Garside of the Department of Zoology, Dalhousie University, Dr P. Colby of the Ontario Ministry of Natural Resources, and Dr D. Alderdice of the Fisheries and Marine Service, Pacific Biological Station for critically reviewing this manuscript; Dr C. R. Sullivan, Department of the Environment for reading the manuscript, and D. Robinson for technical assistance. Special thanks are extended to Dr G. H. Algie of the Sault Ste. Marie Plummer Memorial Public Hospital for providing X-ray facilities.

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Occurrence of Pink Salmon (*Oncorhynchus gorbuscha*) in Lake Huron[1]

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COLLINS, J. J. 1975. Occurrence of pink salmon (*Oncorhynchus gorbuscha*) in Lake Huron. J. Fish. Res. Board Can. 32: 402–404.

Pink salmon (*Oncorhynchus gorbuscha*), which were introduced into Lake Superior in 1956 and have produced eight natural generations there, have now appeared in Lake Huron. The first pink salmon was taken in Ontario waters of Lake Huron in 1971. Successful spawning was observed in one tributary in 1973, indicating that pink salmon could become established in Lake Huron.

COLLINS, J. J. 1975. Occurrence of pink salmon (*Oncorhynchus gorbuscha*) in Lake Huron. J. Fish. Res. Board Can. 32: 402–404.

Le saumon rose (*Oncorhynchus gorbuscha*), introduit dans le lac Supérieur en 1956, où il a produit huit générations naturelles, a maintenant fait son apparition dans le lac Huron. Le premier saumon rose a été capturé dans les eaux ontariennes du lac Huron en 1971. On a observé une ponte réussie dans un tributaire en 1973, ce qui indique que le saumon rose pourrait s'établir en permanence dans le lac Huron.

Received November 15, 1974
Accepted December 6, 1974

Reçu le 15 novembre 1974
Accepté le 6 décembre 1974

PINK salmon (*Oncorhynchus gorbuscha*) were introduced into the Laurentian Great Lakes in June 1956, when the Ontario Department of Lands and Forests (now Ministry of Natural Resources) released about 21,000 fingerlings into the Current River, tributary to Lake Superior and 350 at Pie Island (Nunan 1967). The stock, which originated in the Skeena River, British Columbia. (MacKay 1963), has retained its characteristic 2-yr life cycle, spawning in odd-numbered years. As reviewed by Parsons (1973) adult pink salmon were first reported in Lake Superior by Schumacher and Eddy (1960) and scattered sightings were subsequently reported by Schumacher and Hale (1962) and Moore and Braem (1965). By 1971 the eighth generation was apparently abundant and widespread (Lawrie and Rahrer 1972).

[1]Contribution No. 74-18 of the Ontario Ministry of Natural Resources, Fish and Wildlife Research Branch, Maple, Ont.

Printed in Canada (J3621)
Imprimé au Canada (J3621)

Pink salmon were first observed in Lake Huron in 1969 in the Carp River, a tributary on the upper Michigan Peninsula, where one spent female (427 mm long) was collected (L. Anderson personal communication). The first pink salmon recorded in Ontario waters was captured by a commercial fisherman on August 28, 1971 north of Bigsby Island in the North Channel of Lake Huron. Its identification was confirmed by W. B. Scott of the Royal Ontario Museum, Toronto, Ont. where the specimen has been deposited (ROM Cat. No. 30383). This mature male (fork length 512 mm, weight 1938 g) was considerably larger than Lake Superior pink salmon which ranged in length from 343 to 467 mm (Scott and Crossman 1973).

In 1973 another male pink (fork length 380 mm, weight 570 g) was taken in commercial nets at Burnt Island and in September a female (fork length 384 mm) was caught by an angler in the Mindemoya River, on the south shore of Manitoulin Island. Seven more pink salmon were observed near four redds about 1.7 km upstream from the mouth of that river on October 2. By October 9, spawning had probably terminated as a spent female was the only salmon observed in the area.

Successful spawning — Successful spawning and survival to the alevin stage was confirmed by the retrieval of live eggs and alevins from one redd on November 14, 15, and 27, 1973. Using a Surber sampler and a modified shovel to probe the gravel, the following progeny were obtained:

Date	Live eggs	Dead eggs	Live alevins	Dead alevins	Total progeny
Nov. 14–15	54	4			58
Nov. 27		1	15	34	50

Of 108 eggs and alevins retrieved, 95% had survived to the eyed egg stage of development and 28% of the alevins were alive on November 27. The high rate of alevin mortality was probably induced by repeated probing of the redd. The live eggs collected November 14–15 hatched within 24 h, probably prematurely in response to handling, but the November 27 sample clearly indicated that eggs in the redd had hatched by that date.

Subsequently, 45 alevins (hatched from eggs collected) were cultured in gravel at laboratory water temperatures of about 2 C. Their emergence from the gravel commenced April 8, peaked on April 18, and was complete by April 29. The timing of emergence was within the normal range for pink salmon (Scott and Crossman 1973). However, attempts to capture downstream migrant fry in the Mindemoya River during April were unsuccessful.

Discussion — A number of factors suggest that pink salmon could become established in Lake Huron. Foremost is their apparent success in Lake Superior, the least fertile of the Laurentian Great Lakes (TDS value of 52 ppm, Schelske and Callender [1970]) where pink salmon have apparently overcome the physiological barriers associated with adaptation to fresh water, as have the sea lamprey (*Petromyzon marinus*), alewife (*Alosa pseudoharengus*), and other exotics. This is an unexpected result since Hurley and Woodall (1968) found that pink salmon fry cultured in fresh water, died within 5 wk or less, whereas those cultured in suitable salinity gradients survived well.

Their rapid rate of generation may have facilitated their adjustment to the lacustrine environment. In a relatively short period of 17 yr, the small gene pool has been exposed to selective processes 9 times (eight natural generations). Further, their survival during this period was probably enhanced by the depressed state of predatory lake trout (*Salvelinus namaycush*) populations and the decline of planktivorous lake herring (*Coregonus artedii*) which may be competitors (Lawrie and Rahrer 1972).

As the evidence shows, a preselected stock of pink salmon are attempting to colonize the more fertile waters of Lake Huron. Analogous to the Lake Superior situation, their entry coincides with a period when climax predators and lake herring are not abundant and alewife numbers (another possible competitor) have declined relative to the 1960 decade (Berst and Spangler 1973). Therefore, pink salmon may be regarded as a potential addition to the list of exotics that have become established in Lake Huron.

Acknowledgments — Thanks are due Messrs E. Carlson, G. Purvis (commercial fishermen), and J. Hicks (angler) for the interest they have displayed by turning over specimens to the Ministry.

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Fecundity Comparisons for Various Stocks of Lake Whitefish, *Coregonus clupeaformis*

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Healey, M. C., and C. W. Nicol. 1975. Fecundity comparisons for various stocks of lake whitefish, *Coregonus clupeaformis*. J. Fish. Res. Board Can. 32: 404–407.

We found no significant differences in slope or intercept for the regression of $\log_e$ fecundity on $\log_e$ fork length among samples of whitefish from four lakes near Yellowknife in the Northwest Territories. The equation describing the relationship between fecundity and fork length for these populations was:

$$\text{Fecundity} = 0.0404 \text{ length}^{3.527}_{(\text{cm})}$$

Five other populations for which length–fecundity relationships could be calculated had length exponents ranging from 3.20 to 4.38, suggesting a nonlinear relationship between weight and fecundity. Six of the nine populations as well as four others for which limited data were available all had similar relative fecundities. Fish from Buck Lake in Alberta and from Lake Erie had high relative fecundities while fish from Great Slave Lake had low relative fecundity.

Healey, M. C., and C. W. Nicol. 1975. Fecundity comparisons for various stocks of lake whitefish, *Coregonus clupeaformis*. J. Fish. Res. Board Can. 32: 404–407.

Nous n'avons pas trouvé de différence significative dans la pente ou l'intercepte de la régression du $\log_e$ de la fécondité sur le $\log_e$ de la longueur à la fourche parmi des échantillons de grands corégones de quatre lacs du voisinage de Yellowknife, Territoires du Nord-Ouest. L'équation décrivant la relation entre la fécondité et la longueur à la fourche chez ces populations est:

$$\text{Fécondité} = 0.0404 \text{ longueur}^{3.527}_{(\text{cm})}$$

Cinq autres populations pour lesquelles nous avons pu calculer de relations longueur–fécondité ont des exposants de longueur allant de 3.20 à 4.38, ce qui suggère une relation non linéaire entre le poids et la fécondité. Six des neuf populations, ainsi que quatre autres sur lesquelles nous avons des données limitées, ont toutes la même fécondité relative. Les poissons du lac Buck en Alberta et du lac Erié ont une haute fécondité relative, alors que ceux du Grand lac des Esclaves ont une basse fécondité relative.

Received July 22, 1974
Accepted December 3, 1974

Reçu le 22 juillet 1974
Accepté le 3 décembre 1974

Printed in Canada (J3498)
Imprimé au Canada (J3498)

THERE are scattered references in the literature to the fecundity of lake whitefish (*Coregonus clupeaformis*) (Qadri 1968; Hart 1931; Cucin and Regier 1965; Christie 1963) and some studies in which fecundity is reported for a range of fish sizes (Kennedy 1953; Bidgood 1972; Bajkov 1930; Lawler 1961; Milner 1874). Considering the importance of whitefish in the freshwater fisheries of Canada, however, the amount of published information on their fecundity is small. Little attempt has been made to compare values of this important parameter in different stocks. The purpose of this note is to present some new data on whitefish fecundity and to compare length–fecundity relationships for various stocks.

Methods — Our fecundity estimates are for whitefish from four lakes (Alexie, Baptiste, Chitty, and Drygeese) near Yellowknife in the Northwest Territories. These lakes are oligotrophic, 300–500 ha in area, and have maximum depths of 20–34 m (Healey and Woodall 1973). The lakes are the location of an experimental study into the dynamics of exploited whitefish populations (Healey 1973).

We collected both ovaries from between 40 and 50 maturing females in early October 1972 and late September 1973. The ovaries were preserved in formalin for later estimation of fecundity.

To estimate fecundity we washed the preserved ovaries, at the same time crumbling and kneading them to free the eggs from the ovarian tissue. We used only those ovaries in which there was a clear size differentiation between maturing and residual eggs. We removed as much as possible of the ovarian tissue from the egg mass, then separated and counted a subsample of 3000–5000 maturing eggs. We weighed both the remaining mass of eggs and the subsample to the nearest 10 mg after drying them in air to constant weight. Total fecundity was the ratio of total egg weight to subsample weight multiplied by the number of eggs in the subsample. Errors in fecundity estimation by this technique were $< \pm 2\%$ when compared against total counts (13 comparisons). We found that volumetric and wet weight techniques gave substantially greater errors in estimation (>10%).

Fecundities reported by other authors and reanalyzed in this report were estimated either by a volumetric method (Kennedy 1953) or wet weights (Milner 1874; Lawler 1961; Bidgood 1972) or the method was not reported (Bajkov 1930). W. A. Bond kindly supplied us with some recent data on the fecundity of Great Slave Lake whitefish gained by counting all the eggs in both ovaries.

All fecundities presented in this paper are total numbers of eggs in both ovaries and all lengths are fork lengths. A few authors presented fecundity as eggs per unit weight (Qadri 1968; Cucin and Regier 1965; Christie 1963). These we converted to fecundity of a fish of average length by length–weight relationships reported by the same author.

Results and discussion — Final sample sizes, after we had discarded unusable ovaries, were: Alexie 29, Baptiste 22, Chitty 27, Drygeese 29. The average fork lengths and weights of these fish were: Alexie, 44 cm and 1173 g; Baptiste, 43 cm and 1104 g; Chitty, 41.5 cm and 937 g; Drygeese 47.5 cm and 1404 g. The fish ranged in age from 6+ to 13+ yr.

There were no differences in the slope or intercept of regressions of $\log_e$ fecundity on $\log_e$ fish length among the four lakes. Therefore, we calculated a single regression for the combined samples (Fig. 1A):

$$\text{Fecundity} = 0.0404\ \text{Fork length}^{3.527\ (\text{cm})}$$

This relationship may be considered representative of the fecundity of whitefish from the four lakes, although the scatter of points around the regression is quite wide.

We calculated fecundity on length regressions for nine populations. Five northern populations in lakes near the margin of the Precambrian shield (Alexie, Baptiste, Chitty, Drygeese, Great Slave), three prairie lakes in central Alberta (Pigeon, Buck) and Manitoba (Winnipegosis) and Lake Erie. The length exponents of all populations were greater than 3.0 (3.20–4.38, Fig. 1B) although only the values for Great Slave and Buck Lakes were significantly larger ($P < 0.05$). These exponents are within the range reported by Bagenal (1967) for other fishes. Length exponents greater than 3.0 are indicative of a nonlinear relationship between fecundity and weight. Such relationships are not uncommon (Bagenal 1967). For lake whitefish, the relationship suggests that fecundity per unit weight increases with increasing weight. Among small coregonids in Sweden, however, Toots (1951) noted a drop in fecundity per unit weight with increasing weight.

Relative fecundity differs among the populations for which data is available. Fish of similar size from our lakes, Lake Winnipegosis and Pigeon Lake all have similar fecundities. Whitefish from Great Slave Lake have a low fecundity relative to these lakes while whitefish from Buck Lake and Lake Erie have a high relative fecundity (Fig. 1B).

Observations by authors who presented their data as fecundity per unit of weight (Qadri 1968; Christie 1963; Cucin and Regier 1965) indicate fecundities similar to or less than the fecundity of whitefish from our four lakes (Fig. 1B). Early observations on the fecundity of whitefish from Lake Erie by Milner (1874) also indicate fecundities similar to those in our lakes and consider-

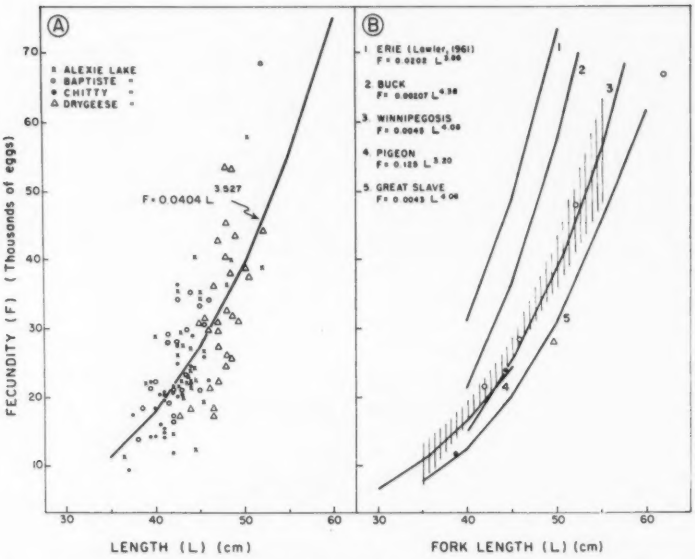

FIG. 1. A, Relationship between fecundity and length for whitefish from Alexie (×), Baptiste (○), Chitty (●) and Drygeese (△) lakes; B, relationship between fecundity and length for different whitefish populations compared with that for Alexie, Baptiste, Chitty, and Drygeese. Shaded area: 95% confidence limits of the regression of fecundity on length for the lakes near Yellowknife. Solid lines: fecundity on length relationships for populations as noted on the figure. Symbols: individual observations for: ○ Lake Erie (Milner 1874); ● Lac la Ronge (Qadri 1968); △ Huron (Cucin and Regier 1965); × Ontario (Christie 1963).

ably less than that observed by Lawler (1961) (Fig. 1B).

Racial or population differences in fecundity have been noted for many fishes (Bagenal 1966, 1967; Baxter 1959; McFadden et al. 1965). Differences in relative fecundity among populations of whitefish are of a similar magnitude to those observed for North Sea herring (Baxter 1959) and plaice (Bagenal 1966). The data do not permit any conclusions as to the cause of differences in fecundity among whitefish populations. Numerous factors, including nutritional state (Scott 1961; McFadden et al. 1965), time of sampling and maturity stage (Healey 1971), and racial characteristics (Bagenal 1966), have been suggested to explain variations in fecundity both within and between fish populations. Considering the wide range of lake types and geographic locations from which the fecundity estimates come, any or all the suggested factors may be operating.

Acknowledgments — Technical support was provided for this study by C. Crocker and K. Rowes. G. B. Ayles criticized a draft of the manuscript. We are grateful to Mr W. Bond for permitting us to use his fecundity data from Great Slave Lake.

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Annual Growth Rates of Mooneye, *Hiodon tergisus*, in the Assiniboine River

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GLENN, C. L. 1975. Annual growth rates of mooneye, *Hiodon tergisus*, in the Assiniboine River. J. Fish. Res. Board Can. 32: 407–410.

Estimates of back calculated lengths at each age for 669 mooneye (*Hiodon tergisus*) from the Assiniboine River indicate that annual length increments decrease progressively with age. Weight increments, calculated from estimates of weights at corresponding lengths, were greatest for mooneye during their fourth year of life. Both sexes of young mooneye had similar growth rates but after age III females grew more rapidly than male fish.

GLENN, C. L. 1975. Annual growth rates of mooneye, *Hiodon tergisus*, in the Assiniboine River. J. Fish. Res. Board Can. 32: 407–410.

Les longueurs à chaque âge, estimées par rétrocalculs, de 669 laquaiches argentées (*Hiodon tergisus*) de la rivière Assiniboine indiquent que l'augmentation annuelle de longueur diminue progressivement avec l'âge. Les augmentations de poids, calculées à partir d'estimés de poids à des longueurs correspondantes, sont à leur maximum, chez la laquaiche argentée, à la quatrième année. Les jeunes laquaiches argentées des deux sexes ont un rythme de croissance semblable, mais passé l'âge III, les femelles croissent plus rapidement que les mâles.

Received August 19, 1974
Accepted December 10, 1974

Reçu le 19 août 1974
Accepté le 10 décembre 1974

MOONEYE, *Hiodon tergisus*, are abundant in the Assiniboine River in Manitoba. According to Scott and Crossman (1973), this population represents one of the more western occurrences in the geographic range for the species. Growth studies of mooneye have been published for eastern populations. Van Oosten (1961) presented data computed from scale dimensions for 130 mooneye aged I–VII captured from Lake Erie during 1927–29. Additional data for mooneye from Lake Erie and Lake St. Clair have been

Printed in Canada (J3529)
Imprimé au Canada (J3529)

published by Johnson (1951) but quantitative growth data on mooneye west of the Great Lakes and on river populations are required. The purpose of this investigation was to calculate annual growth rates from scale criteria for mooneye in the Assiniboine River.

Methods — The mooneye population in the Assiniboine River at Brandon, Man. was sampled at 2-wk intervals between November 17, 1972 and November 2, 1973 with nylon gillnets (stretch mesh measures 38.1, 63.5, and 88.9 mm) placed as bottom sets across the river. Total length and weight of each of the 669 mooneye netted were measured. Adult fish from this sample were sexed by examination of gonads and presence of the lobed anal fin of males. Data from the sexually mature fish were used to compare male and female growth rates.

The scale method, as described by Van Oosten (1929) for lake herring, was used to determine age and growth of the mooneye. Several scales from each fish were selected from the left side anterior to the dorsal fin immediately above the lateral line. A cleaned scale was placed between microscope slides and, with the aid of a microprojector, its magnified image was projected on to a screen. Annuli, recognized by spacing of circuli and anastomosis of circuli in the lateral field, were counted to determine age without reference to fish. Occasionally the annulus was recognized by erosion of circuli in the lateral field, a condition similar to that noted by Martin (1952) for goldeye (*Hiodon alosoides*). More than one scale of each fish was examined if images were unclear or annuli not distinct.

To determine the beginning of the growing season, scales were examined for a peripheral annulus. Scales from 45 mooneye netted on May 28 showed no evidence of new annulus formation. All scale samples from 22 fish of ages I and II collected on June 12 and all 31 scale samples examined from fish netted June 27 (ages I–V) had a distinct peripheral annulus. Consequently, the first week in June was considered the beginning of the growing season and all fish netted after this date were assumed 1 yr older and were assigned to the next age-group.

Estimation of body length at beginning of each growing season was obtained from the measurement along the anterior scale radius between each annulus and scale focus. The total anterior radius of each scale was also measured. Back calculated lengths at annulus formation were then obtained from a body length to scale length ratio with a correction factor as described by Fraser (1920). The correction factor was determined by plotting scale measurements against body lengths and extrapolating a least squares regression line. Mean body lengths were grouped into 1-cm intervals and were plotted against mean anterior scale radii (Fig. 1). The Y intercept of 53 mm was used as the correction factor. Because the straight line gave a good fit ($r = 0.99$), it is assumed that scale growth was proportional to body length after the fish had attained 53 mm. The weight of each fish at annulus formation was established from a length–weight relation (Fig. 2) calculated by grouping the estimated lengths into 1-cm intervals.

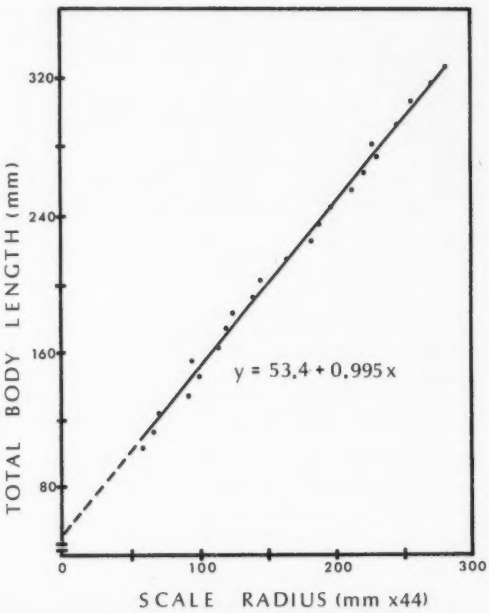

FIG. 1. The relationship between various total length intervals of 669 mooneye (*Hiodon tergisus*) captured from Assiniboine River and the corresponding mean anterior scale radius for each length interval.

Annual increments in length and weight were calculated for each fish. Data from each year-class were combined to obtain mean size estimates at subsequent ages. To compare growth of each sex, annual length increments were calculated separately for the adult male and female mooneye.

Results and discussion — Length at annulus formation was back calculated for 669 mooneye aged I–IX inclusive. The most rapid growth in length occurred during the first year of life, after which annual length increments decreased (Table 1). Mean lengths at comparable ages in different year-classes were relatively constant.

Back calculation of body lengths at annulus formation with a correction factor seemed to be a better procedure than a direct body–scale relationship for mooneye in Assiniboine River. Hile (1970), in his review on methods for calculation of growth in fishes, stated that the intercept value in the body–scale relationship has no morphological significance and may vary with location of

TABLE 1. Mean total lengths (millimeters), estimated mean weights (grams), and annual growth increments of mooneye (*Hiodon tergisus*) netted from the Assiniboine River during 1972–73.

Age	No. of fish	I	II	III	IV	V	VI	VII	VIII	IX
I	181	117								
II	202	126	185							
III	43	119	181	227						
IV	57	111	168	216	250					
V	87	112	170	218	250	272				
VI	77	112	168	215	248	271	286			
VII	17	114	171	219	251	270	286	298		
VIII	2	109	165	208	246	267	285	300	307	
IX	3	107	168	209	253	277	296	310	319	328
Grand mean Length		114±7.7[a]	172±12.4	216±14.8	250±15.3	271±13.2	288±11.6	303±13.1	313±16.4	327±18.1
Weight		14	52	107	169	219	265	309	347	374
Increments										
Length		114	59	46	36	22	17	14	8	9
Weight		14	38	55	62	50	46	44	38	37

[a]Standard deviation.

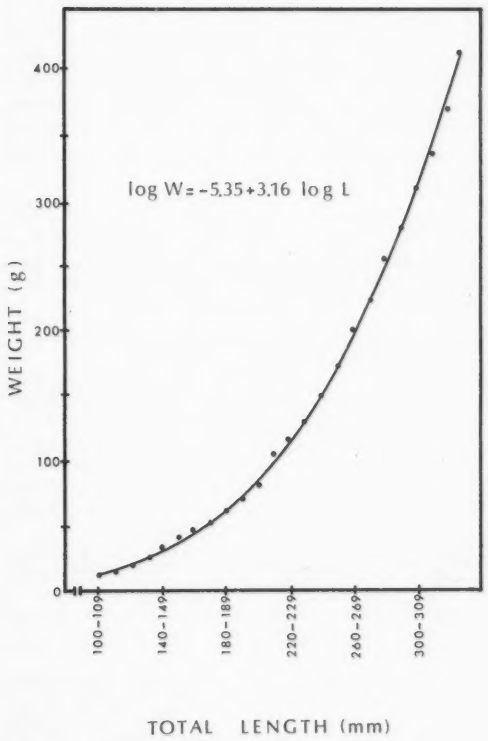

FIG. 2. Length–weight relationships of 669 mooneye from Assiniboine River.

scales used in the calculation. When the intercept value of 53 mm (Fig. 1) was applied as the correction factor, however, estimated lengths at each age approached actual observed lengths at time of annulus formation in 1973. For example, the estimated length of fish age III+ at outer annulus using the correction factor was 227 mm. The observed mean length for 17 fish captured during May (month prior to third peripheral annulus formation) was 235 mm. It was also noted that observed mean lengths of all age-groups sampled during May were larger than corresponding grand mean lengths. Consequently, if the correction factor had not been used, a more biased estimate would have occurred.

The primary cause for the variation in mean lengths at comparable ages for different year-classes is attributed to differences in annual growth rates. The effects of Lee's phenomenon and the failure to recognize annuli on scale samples are believed to produce only minor bias to estimates. Lee's phenomenon, which is the tendency for estimated lengths calculated from scales of older fish to be less than values calculated from younger fish, was noted in ages I–III. Effects from the phenomenon could be reduced in this study by using logarithmic values as described by Fry (1943) but the arithmetic values presented should be subject to only moderate bias. The grand mean lengths calculated for ages I–III may, therefore, be underestimated and the back calculated lengths from younger fish may be more valid. Scale annuli were easily recognized for

TABLE 2. Mean total length (millimeters) and mean estimated weight (grams) at annulus formation for sexually mature mooneye netted from Assiniboine River during 1972–73.

	Length		Weight	
Age	Male	Female	Male	Female
IV (108, 131)[a]	248	252	165	173
V (78, 108)	267	274	209	226
VI (36, 63)	281	291	245	275
VII (8, 14)	291	305	275	318

[a]Numbers of males and females.

young fish but difficulties did occur in scales from fish older than age V. Any errors in identification of annuli for calculation of annual growth in older fish would be complemented by the small sample sizes of the older age-groups. Because several scales were examined if annuli were not distinct, errors caused by incorrect aging are assumed to be minimized.

Mooneye in the Assiniboine River are, in general, smaller than mooneye from Lake Erie and Lake St. Clair. The lengths of respective ages computed from scale dimensions by Van Oosten (1961) for Lake Erie mooneye ranged between 19–38 mm larger within age-groups II–VII. Age I mooneye in the Assiniboine River, however, were 21 mm larger than those from Lake Erie. Johnson (1951) reported that mean lengths of mooneye from Lake Erie and Lake St. Clair ranged from 223 mm at age II to 299 mm at age VI. These mean lengths should not be compared directly with data from the present study because the means were obtained from actual lengths of fish collected during the summer and were not lengths calculated at time of annulus formation.

Annual weight increments increased from age I to IV and then decreased progressively (Table 1). The large weight increment at age IV is the result of both length increment and weight gain per length unit. Figure 2 illustrates that the slope (rate of weight gain per length unit) increased exponentially. Therefore, weight increase per unit length increase of mooneye older than age IV is greater than in younger fish but because these older fish do not have large annual length increments, weight increase is restricted. Conversely, mooneye younger than age III grow rapidly in length but their weight gain per unit length increase is small. Consequently, the large weight increase recorded for mooneye age IV is caused by relatively large length increments between ages III–IV and the relatively large weight gain per length unit.

In the Assiniboine River, most male mooneye become sexually mature in their fourth year of life while females generally mature 1 yr later (unpublished data). Estimates of back calculated lengths at annulus formation from adult fish netted were similar for both sexes between ages I–III. After age III, female mooneye tended to grow more rapidly than males (Table 2). Data from the 230 adult male and 316 adult female fish indicate that females are approximately 4, 7, 10, 14 mm longer at ages IV–VII, respectively. The decrease in the ratio of male to female fish captured and the absence of male fish ages VIII and IX are presumably due to a lower female mortality rate. Scott and Crossman (1973) also reported that male mooneye have a shorter life span than females.

The relatively small number of age III mooneye captured may be due to differences in year-class strength rather than in net selectivity. Only 17 fish age III were captured between November 7 and May 28 (first week of June considered time of age change). Eight mooneye of age IV were captured between June 12 and November 2. Because 49 fish in age-group IV were netted during winter and spring and because mesh sizes were constant throughout the year, the mooneye of this age-class should have been of a size which permitted capture. No attempt can be made to explain the relative strengths of the individual year-classes.

Acknowledgments — I thank Drs R. F. C. Smith and R. K. Springer of Brandon University for suggestions during preparation of the manuscript. I also thank Messrs G. Moran, B. Bruce, and J. Mitchell for their assistance in collection of specimens.

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Use of Sodium Pentachlorophenate and Dehydroabietic Acid as Reference Toxicants for Salmonid Bioassays

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DAVIS, J. C., AND R. A. W. HOOS. 1975. Use of sodium pentachlorophenate and dehydroabietic acid as reference toxicants for salmonid bioassays. J. Fish. Res. Board Can. 32: 411–416.

Results of an interlaboratory bioassay standardization exercise involving seven independent fish bioassay laboratories in British Columbia are presented. Toxicity of standard solutions of sodium pentachlorophenate and dehydroabietic acid was determined in freshwater static bioassays with underyearling rainbow trout (*Salmo gairdneri*), coho salmon (*Oncorhynchus kisutch*), and sockeye (*Oncorhynchus nerka*) salmon. Reported LC50s for the three species ranged from 37 to 130 ppb for sodium pentachlorophenate and 1.03–2.14 ppm for dehydroabietic acid. In general, test results for individual species and toxicants were fairly consistent and major disparities could be explained in some instances by variations in physical and chemical characteristics of the bioassay such as water temperature, hardness, or pH. Fish size and condition factor appeared to have little effect on apparent toxicity over the ranges tested.

The usefulness of reference toxicants for standardizing bioassays is emphasized and suggestions are made for improved test procedures and increasing accuracy of results. Guidelines for toxicity tests should include the use of reference toxicants as a means of standardizing bioassay results.

DAVIS, J. C., AND R. A. W. HOOS. 1975. Use of sodium pentachlorophenate and dehydroabietic acid as reference toxicants for salmonid bioassays. J. Fish. Res. Board Can. 32: 411–416.

Les auteurs présentent les résultats d'un exercice de standardisation d'essais biologiques entre laboratoires, exercice impliquant sept laboratoires indépendants d'essais biologiques des poissons en Colombie-Britannique. La toxicité des solutions-étalons de pentachlorophénate de sodium et d'acide déshydroabiétique a été déterminée lors d'analyses biologiques statiques en eau douce avec des alevins de moins d'un an de truite arc-en-ciel (*Salmo gairdneri*), saumon coho (*Oncorhynchus kisutch*) et saumon nerka (*Oncorhynchus nerka*). Les CL50 rapportées pour les trois espèces vont de 3.7 à 130 ppb pour le pentachlorophénate et de 1.03 à 2.14 pour l'acide déshydroabiétique. En général, les résultats des essais sont assez uniformes pour les espèces et les toxiques individuels, et les différences majeures peuvent être expliquées dans certains cas par des variations dans les caractéristiques physiques et chimiques de l'essai biologique, telles que la température, la dureté et le pH de l'eau. La taille et la condition d'embonpoint des poissons semblent avoir peu d'effet sur la toxicité apparente, dans les limites testées.

Les auteurs soulignent l'utilité de toxiques de référence pour la standardisation des essais biologiques et suggèrent des moyens d'améliorer le protocole des essais et d'augmenter la précision des résultats. Les directives relatives aux essais de toxicité devraient inclure les toxiques de référence comme moyen de standardiser les résultats d'essais biologiques.

Received May 23, 1974
Accepted November 29, 1974

Reçu le 23 mai 1974
Accepté le 29 novembre 1974

THIS study was a bioassay standardization exercise involving seven laboratories, both government and private, in the lower mainland/coastal British Columbia area. Each test laboratory was provided with identical reference toxicant materials and a comprehensive set of guidelines for conducting standard 96-h static bioassays so that results obtained by various laboratories could be compared.

Considerable interest exists in methods of stan-

Printed in Canada (J3437)
Imprimé au Canada (J3437)

TABLE 1. *A list of the laboratories involved in the standardization exercise, their water source and its prebioassay treatment.*

Laboratory	Address	Water source	Water treatment
Environmental Protection Service	4160 Marine Dr. West Vancouver, B.C.	Vancouver City Water Cleveland Dam	none
B.C. Research	3650 Westbrook Cres. Vancouver, B.C.	Vancouver City Water Cleveland Dam	dechlorinated
Fisheries Research and Development	4160 Marine Dr. West Vancouver, B.C.	Well Water — from site	aeration
Pollutech Pollution Advisory Services Ltd.	104 Charles St. North Vancouver, B.C.	North Vancouver City Water — Seymour River	dechlorinated
Beak Consultants Ltd.	124 S.E. Marine Dr. Vancouver, B.C.	Vancouver City Water Cleveland Dam	dechlorinated
International Pacific Salmon Fisheries Commission	Sweltzer Creek Station Cultus Lake, B.C.	Cultus Lake Water	none
Province of B.C. Pollution Control Branch	Fish and Wildlife Lab. 1019 Wharf Street Victoria, B.C.	Victoria City Water Sooke Lake	1 megohm deionized

dardization for bioassays. Recently, federal legislation was promulgated for regulating the toxicity of pulp and paper wastes and petroleum refinery wastes and specific legislation for other toxicants is forthcoming or pending. These regulations propose effluent toxicity standards to be applied uniformly nationwide for each type of pollutant. The inclusion of reference toxicant tests, along with effluent toxicity tests, could improve the quality of the test procedure for comparative purposes. So far, use of reference toxicants for bioassay standardization appears rare and has been applied only to testing oil dispersants (LaRoche et al. 1970) and pesticides (Marking 1966).

Methods — Details of the location and water characteristics for each laboratory are given in Table 1.

Samples of two reference toxicants were distributed to each laboratory within a 24-h period accompanied by a detailed set of instructions. One stock solution containing 14,418 ppb sodium pentachlorophenate (NaPcP) was prepared from Reagent Grade 2, 3, 4, 5, 6-pentachlorophenol (MW 266.36) by addition to NaOH, according to the procedure of Alderdice (1963). A second stock solution of 6667 ppm dehydroabietic acid (DHAA), prepared from heat disproportionated pine rosin (Hercules — 731 D, Hercules Inc., Pine and Paper Chemicals Dept. Wilmington, Delaware, U.S.A.) according to the procedure of Halbrook and Lawrence (1966), was also provided. Dehydroabietic acid is a resin acid derived from wood and is a major toxicant present in wood processing wastes. Sodium pentachlorophenate is a widely used wood preservative.

The bioassay instructions adhered as closely as was practical to federal bioassay guidelines for pulp and paper wastes. Solution pH was adjusted prior to the test to 7.0 ± 0.1 using NaOH and HCl and was monitored throughout the test along with temperature and dissolved oxygen. Temperature and hardness were set equivalent to fish holding conditions for the test. Consultation with all laboratories locally indicated that it was possible to conduct the tests at 10 ± 2 C. These test conditions and procedures were based upon best available knowledge and practicality after joint discussion with the participants.

Replicate 96-h static bioassays in fresh water were conducted using five toxicant concentrations and two controls run simultaneously. Test fish were 1–3 g rainbow trout (*Salmo gairdneri*), coho salmon (*Oncorhynchus kisutch*), or sockeye salmon (*Oncorhynchus nerka*) according to the stock availability at each laboratory. No attempt was made to standardize the source of test fish. Fish loading density in bioassays was specified as 0.5 g/liter over the 4-day period and 10 fish were to be used per tank unless loading density requirements necessitated more tanks with less fish per tank. Tests were done in 30 liters of aerated solution (>90% O_2 saturation) in covered tanks.

Bioassays were observed at 30 min, 2, 6, 24, 72, and 96 h after the beginning of the test and mortality was recorded as time to cessation of all movement by individual fish. Cumulative percentage mortality at each toxicant concentration was plotted on log-probit paper to estimate the time of 50% mortality (LT50). The incipient 96 h LC50 was estimated in two ways. The first method involved plotting percentage mortality at 96 h for each concentration on log-probit paper and reading the estimated concentration lethal to 50% of the fish from the graph (Sprague 1969). If the data were sufficient, a second determination of 96 h LC50 was carried out which involved fitting a

line to log-probit data, expressed as concentration vs. percent dead at 96 h, by the nomographic procedures of Litchfield and Wilcoxon (1949). This latter procedure estimates the line of best fit by minimizing its Chi^2 value and calculates 95% confidence limits for the LC50. (Both procedures follow the same method, although the second is a more accurate refinement of the first.) Slope function, or S, is also calculated so that other workers can reproduce the line, if desired. The LC50s produced by the above procedures are defined as the concentration lethal to 50% of the fish in 96 h under the conditions of the test.

Results — Results of the bioassays with both reference toxicants, as presented by the seven laboratories, using the three species, are summarized in Table 2. The laboratories are not identified but are coded in a random fashion. Pertinent data including fish loading density, temperature, pH, water hardness, fish weight, and fish length are reported. Fish condition factor was calculated from $wt/leng.^3$ where:

wt = weight in g, leng. = length in cm

(Lagler 1956).

Two estimates of LC50 appear in Table 2. The nomographic LC50, complete with confidence limits, allows comparison of different LC50s (LC50s can be considered different if their respective 95% confidence limits do not overlap — J. B. Sprague personal communication). Clearly, there are both similarities and disparities between LC50s determined for the various laboratories.

Discussion — The results and pertinent information provided by each laboratory in Table 2 indicate that LC50s, as compared by examining the confidence limits, are moderately comparable. For example, results for rainbow trout in sodium pentachlorophenate indicated that LC50s obtained by laboratories E, D, and B were comparable. Similarly, those for laboratories C and A were comparable but significantly lower than those for laboratories E, D, and B. The apparently greater sensitivity of fish used by laboratories C and A could be due, in part, to the low initial solution pH present for labortory A or to some factor not evident in the results such as general fish health, state of activity etc. Alderdice (1963) reported that the dissociation ratio of sodium pentachlorophenate to pentachlorophenol is pH dependent. At pH's above 7.4 it is 99% ionized. Progressively more of the more toxic phenolic moiety is present as pH drops below 7.4. Similarly, the differences in sockeye underyearling LC50s in dehydroabietic acid may be related to the disparity of water hardness and test temperature reported by the two laboratories testing that substance.

It should be emphasized that LC50s reported for dehydroabietic acid may be somewhat high as they represent theoretical concentrations in solution. G. M. Kruzynski (personal communication) found that in continuous flow bioassays an average of approximately 89% of dehydroabietic acid was actually present in solution as determined by gas chromatography of water samples in the theoretical concentration range 2.0–0.63 ppm. He also found that dehydroabietic acid concentration in static bioassays declines considerably with time. For this reason, this substance may be a poor choice as a reference toxicant unless gas-chromatographic procedures are employed to accurately determine the actual solution concentration available during the bioassay.

No apparent trend was discernable between fish size or condition factor for the groups used nor was there a large difference in species sensitivity. Lack of a condition factor effect is in general agreement with the findings of Davis and Mason (1973) who reported that a condition factor range of 0.8–1.4 g/cm^3 did not affect toxicity test results with bleached kraft pulpmill effluent.

An observation made by many of the participants was that the data were weakened considerably by a lack of frequent observations, particularly in the early part (first 24 h) of the bioassay. The choice of infrequent observations was based partly on economic and personnel considerations of the private companies participating.

Sprague (1973) suggested the following convenient set of observation times:

Initial hour	Min: 15, 30, 60
1st day and a half	Hours: 2, 4, 8, 14 ± 2, 24, 33 ± 3
2nd day, etc.	*Days: 2, 3, 4 and daily* beyond this

He points out that if a test is begun in mid-morning all observations can be made during normal working hours, with only one observation needed late in the evening of the first day.

Data quality was also influenced by the choice of test concentrations used for bioassays. For example, no fish would die in 96 h in a given concentration while at the next concentration mortality could be at, or close to, 100% in 96 h. With this sort of data, LC50 can only be crudely estimated by log-probit methods owing to the paucity of points available for plotting. In addition, use of the nomographic procedure of Litchfield and Wilcoxon (1949), is not possible as a plot of partial mortality (probit scale) vs. concentration (log scale) is required.

TABLE 2. Results of static bioassays and summary of test conditions reported by the various laboratories coded in a random fashion. LC50s were calculated in two ways, as a log-probit estimate or as an estimate resulting from a nomographic analysis yielding 95% confidence limits for the LC50 (upper and lower limits in brackets). The slope function, S, for the nomographic analysis is given. Fish condition factor, C.F., was calculated using the mean fish weight and length given in the table for each test.

Lab. code	LC50 Log-probit estimate	LC50 Nomographic calc. + 95% CL	S	Loading density (g/l)	x̄ initial temp-C	x̄ initial pH	x̄ hardness ppm	x̄ wt g	x̄ length cm	C.F. Wt/L³
		rainbow trout — ppb sodium pentachlorophenate								
E	92	98(87.5, 109.8)	1.136	0.52	12.0	6.96	51.5	2.84	5.98	1.33
D	96	96(90.1, 102.2)	1.109	0.42	11.54	7.0	47.0	0.87	4.28	1.11
C	50	50(29.8, 84.0)	1.99	0.48	11.0±0.1	7.0±0.1	5–6	1.90	5.80	0.97
B	100	106(88.5, 127.0)	1.331	≃0.50	11.5	7.02	5.0	1.52	5.24	1.06
A	48	47(32.9, 67.2)	1.732	≃0.50	10.04	5.7	4.0	1.39	4.84	1.23
F	75	–[a]	–	0.58	10.0±1.0	7.0±0.1	10.0	1.31	–	–
		rainbow trout — ppm dehydroabietic acid								
E	1.03	1.06(0.99, 1.14)	1.116	0.54	10.94	6.98	23.0	3.33	6.32	1.31
D	1.05	–	–	0.57	12.25	6.97	47.0	1.26	–	–
C	1.22	–	–	0.48	11.0±0.1	7.0±0.1	5–6	1.90	5.80	0.97
B	1.74	1.85(1.62, 2.11)	1.237	≃0.50	11.5	6.92	5.0	1.40	5.32	0.93
A	1.18	1.20(1.03, 1.39)	1.118	≃0.50	9.3	9.0	4.0	1.67	4.96	1.37
F	1.25	–	–	0.57	10±1.0	7.0±0.1	10.0	1.29	–	–
		coho salmon — ppb sodium peniachlorophenate								
E	96	92(79.3, 106.7)	1.12	0.76	10.25	7.01	14.7	4.61	7.40	1.14
C	37	31.8(24.9, 40.6)	1.317	0.52	11.0±0.1	7.01±0.1	5–6	2.14	5.80	1.10
		coho salmon — ppm dehydroabietic acid								
E	1.38	–	–	0.69	10.62	6.97	7.0	4.18	6.24	1.72
C	1.76	1.75(1.56, 1.96)	1.134	0.52	11.0±0.1	7.1±0.1	5–6	2.14	5.80	1.10
		sockeye salmon — ppm dehydroabietic acid								
D	1.38	–	–	0.46	11.56	7.07	47.0	1.70	–	–
G	2.14	2.23(2.03, 2.45)	1.114	0.45	7.5	7.7	85.0	1.38	4.92	1.16
		sockeye salmon — ppb sodium pentachlorophenate								
D	50	50(40.3, 62.0)	1.281	0.56	12.5	7.19	47.0	0.68	–	–
G	130	130(119, 142)	1.096	0.46	7.5	7.70	85.0	1.38	5.05	1.07

[a]Data insufficient for calculation of confidence limits.

The data could therefore have been strengthened by inclusion of more replicates of intermediate toxicant concentrations, particularly those concentrations close to the LC50 value. It seems wise to suggest that at least three test concentrations be used between the concentration where little or no death occurs, and where all or most of the fish die within 24–48 h. Standard Methods (1971) gives useful tables for preparing dilutions of test solutions.

To our knowledge, a little work has been done using reference toxicants as a means of standardizing bioassay procedures. Marking (1966) evaluated *p, p'*-DDT as a reference toxicant for bioassay use. LaRoche et al. (1970) suggest "the use of a reference toxicant is desirable as a link between the findings of different investigators, as an internal standard to compare the relative toxicities of substances, and as a measure of condition of test organisms." The Canadian Environmental Protection Service's Report (EPS 1-EE-73-1, 1973) recommends use of reagent grade dodecyl sodium sulphate (also known as DSS or sodium lauryl sulfate) as being "rapid, nonselective and consistent in its toxicity to aquatic animals." That report requires that the median lethal DSS concentration be determined each time an oil dispersant bioassay is conducted. The report suggests that the median lethal DSS concentration may be in the general range of 1–10 mg/liter. Apparently one problem associated with the use of DSS is its tendency to foam when used in continuous-flow bioassays (D. G. Alexander personal communication).

Alderdice (1963) selected sodium pentachlorophenate, as being a suitable "toxic stressor" for fish after screening thirty-five chemicals of considerable toxicity to fish. His selection was based on the following properties of sodium pentachlorophenate:

"high solubility in water
virtually complete ionization in solution at pH 7.0 or higher
stability in solution at pH 7.0–8.5
availability as a dry solid of high purity
facility of quantitative determination
toxicity to fish (in low concentrations)"

These criteria would seem to be excellent guidelines for the selection of other reference toxicants although the necessity for complete ionization is not applicable to toxic organics such as pulpmill waste constituents. Ideally, a group of substances should be available, selected from a list of major toxicants such as pesticides, detergents, heavy metals, wood-processing effluent, petrochemical wastes and other pollutants. Equally useful might be some stressor such as lethal temperature or salinity. Howard (1973) used lethal temperature as a stressor to evaluate the response of fish to kraft pulpmill waste, copper, zinc, and phenol.

It should be emphasized that bioassay results can be affected by many factors including fish loading density, fish size and age, photoperiod, fish handling and exposure to visual stress, temperature, acclimation, water hardness and pH, aeration, and others. Great care must be exercised in comparing bioassay results to insure that a meaningful comparison is possible. Guidelines and pertinent references for conducting bioassays are found in Sprague (1969), Standard Methods (1971), and Davis and Mason (1973).

In summary, the use of reference toxicants, such as sodium pentachlorophenate and dehydroabietic acid, yields useful information on the comparability of fish toxicity tests by independent laboratories. Fairly consistent results can be obtained provided a detailed set of guidelines is provided and criteria such as fish loading density, acclimation, test temperature, test tank size, statistical procedures, frequency of observation, water pH, and volume of test solution are specified. Reference toxicants provide a means of detecting large differences in sensitivity of fish stocks to toxicants that may be the result of the state of health of the stock or some variation in one of the above bioassay conditions. It is probably naive, however, to expect that any two stocks or bioassay tests will yield identical results owing to the tremendous scope of biological and environmental variation. Indeed, the study of Marking (1966), involving toxicity of *p, p'*-DDT of 19 species of freshwater fish, failed to demonstrate consistency of toxicity for many of these species. The usefulness of reference toxicants therefore, may be confined to detecting large differences in sensitivity of test fish, which are likely to have a serious effect on bioassay results. Guidelines for toxicity tests should include the use of reference toxicants as a means of standardizing bioassay results.

Acknowledgments — In reality this paper belongs to all the participating laboratories who were involved in the study. The assistance of Mr W. N. Holman, Mrs D. Price, and Mr I Shand in preparing this report is appreciated, as are the useful comments of Drs D. Alderdice, T. E. Howard, I. H. Rogers, J. B. Sprague, and M. Waldichuk. Drs D. F. Alderdice and I. H. Rogers helped greatly in the choice and preparation of the stock toxicant solutions.

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Perfusion Fixation of Whole Fish for Electron Microscopy

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HINTON, D. E. 1975. Perfusion fixation of whole fish for electron microscopy. J. Fish. Res. Board Can. 32: 416–422.

A method for obtaining uniform fixation of whole bodies of fish for electron microscopic study is described. The procedure employs vascular perfusion of aldehyde fixatives through the proximal aorta of anesthetized fish. Light microscopic examination of semi-thin (0.5–1 μ) sections of Epon embedded material, stained with toluidine blue, showed near to complete removal of blood cells from vessels of gill, bone, skin, skeletal muscle, heart, exocrine pancreas, mesentery, glomerulus, and brain. Electron microscopic examination of liver and kidney, fixed by whole body perfusion, showed excellent preservation of cell ultrastructure.

HINTON, D. E. 1975. Perfusion fixation of whole fish for electron microscopy. J. Fish. Res. Board Can. 32: 416–422.

L'auteur décrit une méthode de fixation uniforme de corps entiers de poissons pour étude au microscope électronique. Il s'agit d'une perfusion vasculaire d'aldéhydes fixateurs par l'aorte proximale de poissons anesthésiés. L'examen au microscope photonique de coupes semi-minces (0.5–1 μ) de matériel enrobé dans de l'Epon, coloré au bleu de toluène, indique l'élimination presque complète de cellules sanguines des vaisseaux de la branchie, de l'os, de la peau, du muscle squelettique, du coeur, du pancréas exocrine, du mésentère, du glomérule et du cerveau. L'examen au microscope électronique du foie et du rein, fixés selon le procédé de perfusion du corps entier, révèle une excellente préservation de l'ultra-structure cellullaire.

Received June 24, 1974
Accepted December 10, 1974

Reçu le 24 juin, 1974
Accepté le 10 décembre 1974

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Printed in Canada (J3472)
Imprimé au Canada (J3472)

THIS report describes a method whereby uniform fixation of entire fish for electron microscopy is possible with a single perfusion. The technique, employing vascular perfusion of aldehyde fixatives, is easily adapted to field as well as laboratory use. Since the early papers on vascular perfusion techniques (Palay 1960; Palay et al. 1962), which described fixation of brain in goldfish, no use of the technique in subsequent fish studies has been reported. Perfusion fixation of single organs such as kidney in rat (Maunsbach 1966; Griffith et al. 1967) and liver in mouse (Sjöstrand 1967) has given superior preservation of fine structural detail. Recent studies on chronic sublethal injury in animal models indicate that alterations are commonly encountered at the subcellular (i.e. organelle) level (Arstila et al. 1974). Consistently uniform fixation of tissues for electron microscopic study would improve the capability of assessing sublethal responses of fishes to environmental toxins and stress.

Fish between 85 and 550 g in weight and 18–45 cm in length were perfused successfully with the described equipment. Species with compressed (channel catfish, *Ictalurus punctatus*) and depressed (striped bass, *Morone saxatilis*) body forms were used.

Procedure for fixation — For anesthetic, prepare a fresh solution of tricaine methane sulfonate (M.S. 222) in water to a final concentration of 1:4000 (McFarland and Klontz 1969). Expose the fish to the anesthetic for 5–10 min, or until it ceases all but opercular movement, loses equilibrium, and is insensitive to touch. Soak 5- × 5-cm gauze sponges in the anesthetic solution and position them under the opercula and over the gill arches. Fill an applicator bottle of 500-ml capacity with the anesthetic solution.

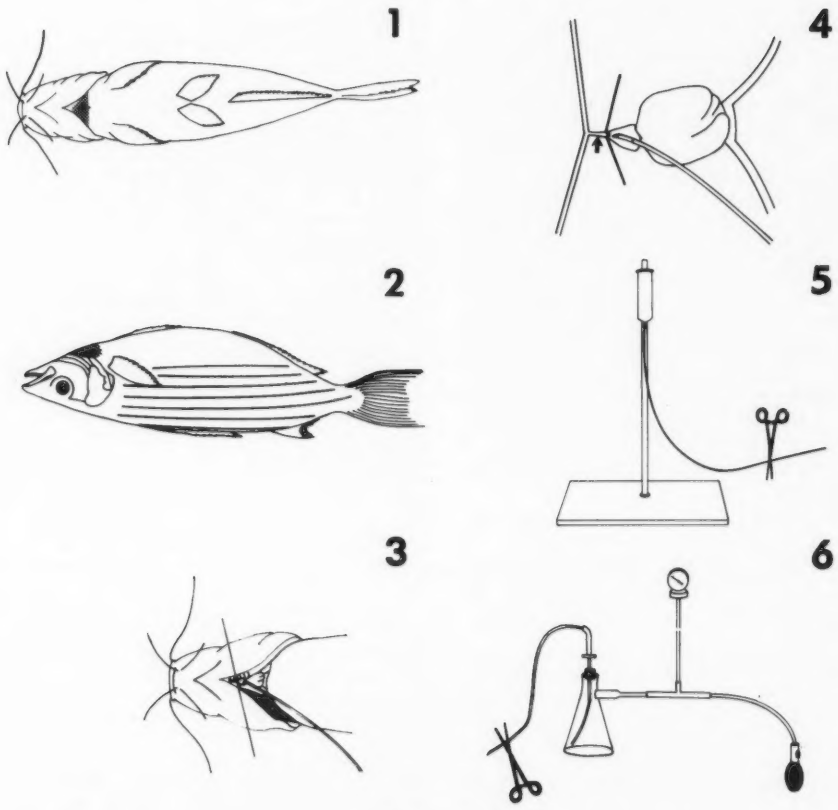
Remove the fish from the water and place in the supine position if it has a depressed body form (Fig. 1), or on its side with the dorsal aspect toward you, if the fish has a compressed body form (Fig. 2), within a reinforced trough.

For the surgical area, if the fish has a depressed body form (Fig. 1), inscribe a triangle, making the sides by percutaneous incisions along the inferomedial body wall near both opercula and the transverse cut by a similar incision just anterior to the bases of the pectoral fins. Use a scalpel and blunt forceps to reflect the skin from the surgical area, and reflect the skeletal muscle to expose the parietal pericardium. Remove this layer to uncover the bulbus arteriosus of the aorta. Enlarge the triangle and expose the heart and aorta (Fig. 3) by placing heavy surgical scissors transverse and superficial to the aorta and cutting away the cranial apex of the pectoral girdle.

For the surgical area, if the fish has compressed body form (Fig. 2), abduct the operculum and underlying gill arches on the side nearest you. This will expose a subtriangular area with transparent skin and little or no scales. To uncover the heart and aorta, make an incision in a vertical plane, from the scale margin along a line about 1 cm in front of the pectoral fin to the inferior margin of the transparent skin. Make a second incision in a horizontal plane along the inferior margin of the transparent skin from the vertical incision diagonally to the ventral midline. Make a third incision along the superior margin of the scale-free area to connect with the other incisions. Reflect the skin and skeletal muscle from this region to expose the lateral margin of the parietal pericardium, behind which the arota should be visible. Enlarge the area by removing adjacent skeletal muscle at the midline of the body. Allow adequate exposure of the aorta and heart without causing extensive trauma to gills and to vessels supplying the body wall and liver. Position applicator bottle and squeeze so that a continuous flow is introduced over the gills. To begin the perfusion, pass a moist cotton thread dorsad to the aorta and tie this in a loose knot between the bulbus arteriosus and the first branchial arch artery. Grasp the lateral margin of the bulbus arteriosus with blunt forceps and, using scissors, make a small opening in the wall of the vessel. Immediately, insert a cannula of polyethylene into the aorta to a point distad to the thread (Fig. 3, 4). Begin the perfusion immediately and tie the thread securely around the vessel and cannula to prevent loss of perfusate from the vessel aperture. Next, make a transverse incision through the ventricular wall to permit escape of blood from the vascular system upon displacement of the fixative. Maintain sufficient pressure (see below) to give a flow rate of 10 ml/min. Perfuse a total of 20–100 ml of fixative per fish, according to size, with larger fish receiving a greater volume.

For perfusion, use either of two types of devices. A gravity feed type, the simpler device (Fig. 5), comprises a ring stand, a 50-ml syringe (perfusate reservoir) and an 18-gauge needle over which polyethylene tubing (internal diameter 0.1 cm, external diameter 0.2 cm) is fitted. Regulate perfusion pressure (60–100 mm Hg) by adjusting the height of the syringe. In general, a height of 1–2 m allows sufficient pressure.

A pressure-pump device (Fig. 6) comprises a 250–ml suction flask with a side arm; a nylon T-shaped tube; a rubber hand pump with an air release valve; a rubber stopper, and rubber tubing; a pressure gauge; polyethylene tubing; a 13- and an 18-gauge needle; and a hemostat. To assemble this device, fit rubber tubing over the side arm of the suction flask. Connect the free end of the tubing to the nylon T-shaped tube. Connect the other ends of the T, with rubber tubing, to the hand pump and to the pressure gauge. Push the shaft of the 13-gauge-needle through the rubber stopper. Fit polyethylene tubing over the tip of the needle and extend it to the floor of the suction flask. With a 5-cm length of rubber tubing connect the base of the 13-gauge needle with the 18-gauge needle and insert the tip of the latter into the polyethylene cannula. Fill the perfusion flask with 50–100 ml of fixative. Force the fixative through the entire tubing to remove all air. Next, use hemostat clamps to occlude the cannula until the perfusion is begun. Before inserting the



FIGURES 1–6

FIG. 1. Position for perfusion of fish with compressed body form. Hatched triangle overlies area for surgical approach; FIG. 2. Position for perfusion of fish with depressed body form. Hatched triangle overlies area for surgical approach; FIG. 3. Superficial and deep structures included in whole body perfusion, from right to left, heart, proximal aorta with cannula inserted, ligature of suture, more distal aorta and initial branchial arch artery. Black, net-like material is gauze sponge placed over gill for administration of anesthetic; FIG. 4. Position of cannula within aorta, arrow points to first branchial arch artery. Black line represents cotton suture; FIG. 5. Gravity feed perfusion apparatus; FIG. 6. Pressure pump perfusion apparatus.

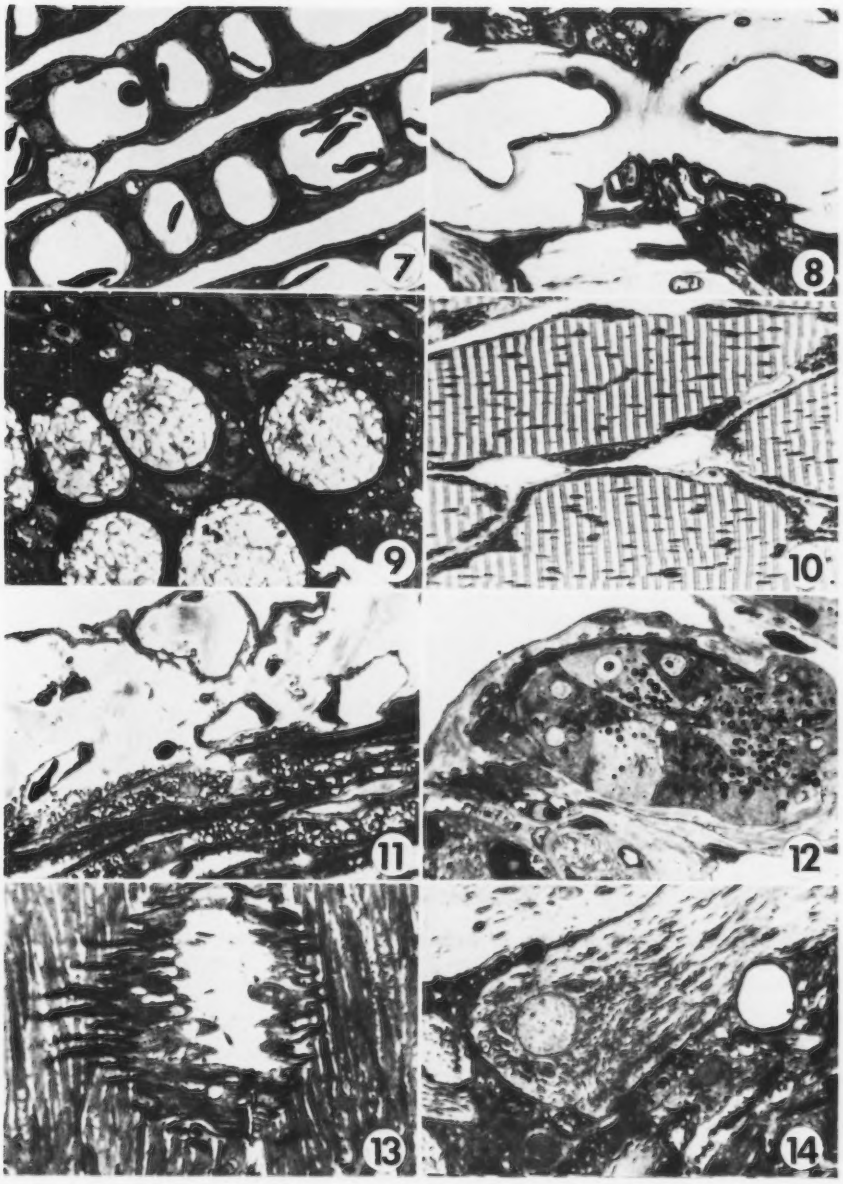
cannula, have a sufficient head of pressure (100 mm Hg) so that perfusion will begin immediately upon release of the clamp.

After the perfusion, cover fish with moist towels and allow 30–60 min for in situ fixation. Recommended fixative solutions are glutaraldehyde (2–4%), formaldehyde–glutaraldehyde (2%, 2%, respectively) (McDowell and Trump 1974), and paraformaldehyde–glutaraldehyde. Preferably, use a buffer that is ⅓ the osmolality of the fish serum (Bone and Ryan 1972; Penttila et al. 1974). Both cacodylate and phosphate buffers (pH 7.4) were satisfactory.

Evaluate fixation, prior to processing for microscopy, by the following criteria. A generalized pale appearance of viscera indicates uniform removal of blood by the fixative solution. Yellowish coloration in glutaraldehyde fixed tissues indicates the extent of penetration by the fixative, especially in mesenteries, skeletal muscle, and nonpigmented areas of ventral body wall. Well-fixed tissues are firm and cut easily with a razor blade.

Occasionally, uneven perfusion due to focal obstruction of the capillary bed occurs in liver and, to a lesser extent, kidney and spleen. As such material is marginally fixed, select the best areas by observing small pieces under the light microscope. If such tissues have patent lumens (kidney) or uniform appearance (liver and spleen) they may be carried through the subsequent procedures.

The subsequent procedures proved successful and



FIGURES 7–14. Light micrographs of tissues from channel catfish (*Ictaluras punctatus*) fixed with 4% glutaraldehyde in cacodylate buffer, pH 7.4 and stained with toluidine blue. ×2000.

FIG. 7. Secondary lamellae of gill, showing capillaries, respiratory epithelium, pillar cells, and mucous cell; FIG. 8. Two blood vessels and adjacent neural tissue in bone of semicircular canal; FIG. 9. Epidermis from lateral aspect of tail; numerous epithelial cells separated by occasional larger cells containing much mucus; FIG. 10. Striated muscle from tail region showing capillaries free of blood cells; FIG. 11. Epicardium (*top*) and myocardium (*bottom*) of ventricular wall; FIG. 12. Mesentery including exocrine pancreas; FIG. 13. Medium sized artery from mesentery; FIG. 14. Large neuron in cerebellum.

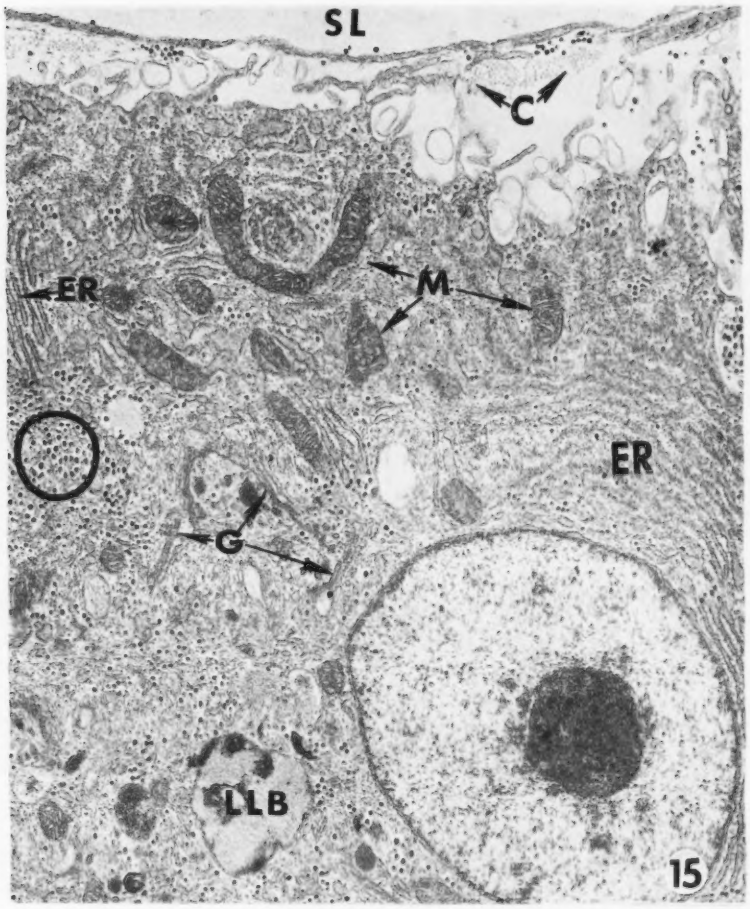


FIG. 15. Electron micrograph of hepatocyte of channel catfish fixed with 4% glutaraldehyde. ×19,000. Phillips 200 electron microscope. SL, sinusoid lumen; C, collagen; M, mitochondria; G, Golgi apparatus; ER, endoplasmic reticulum; LLB, lysosome-like body; circle, glycogen.

are recommended. Mince tissues into 0.5–1 mm pieces and postfix in osmium (Millonig 1961). Dehydrate in graded ethanol solutions, embed in Epon (Luft 1961), and section. Stain thin sections with uranyl acetate (Watson 1958) and lead citrate (Venable and Coggeshall 1965) and view under electron microscope. For correlation between light and electron microscopic observations, stain semithin sections (0.5–1 μ) of Epon embedded material with toluidine blue (Trump et al. 1961) and view under a light microscope.

Merits of the technique — This method of fixation results in excellent preservation of structural detail at both the light (Fig. 7–14) and electron microscopic (Fig. 15, 16) levels. Delicate and hard tissues appear to be equally well fixed. Penetration of fixative from blood vessels into surrounding tissue is evidenced by good fixation of cells some distance removed from their vascular supply. Tissues from inaccessible sites such as pituitary and head kidney are easily fixed in a single procedure.

Ventricular wall of heart revealed differences in quality of fixation with epicardium showing better fixation than the more deeply placed myocardium which revealed extensive vacuolation

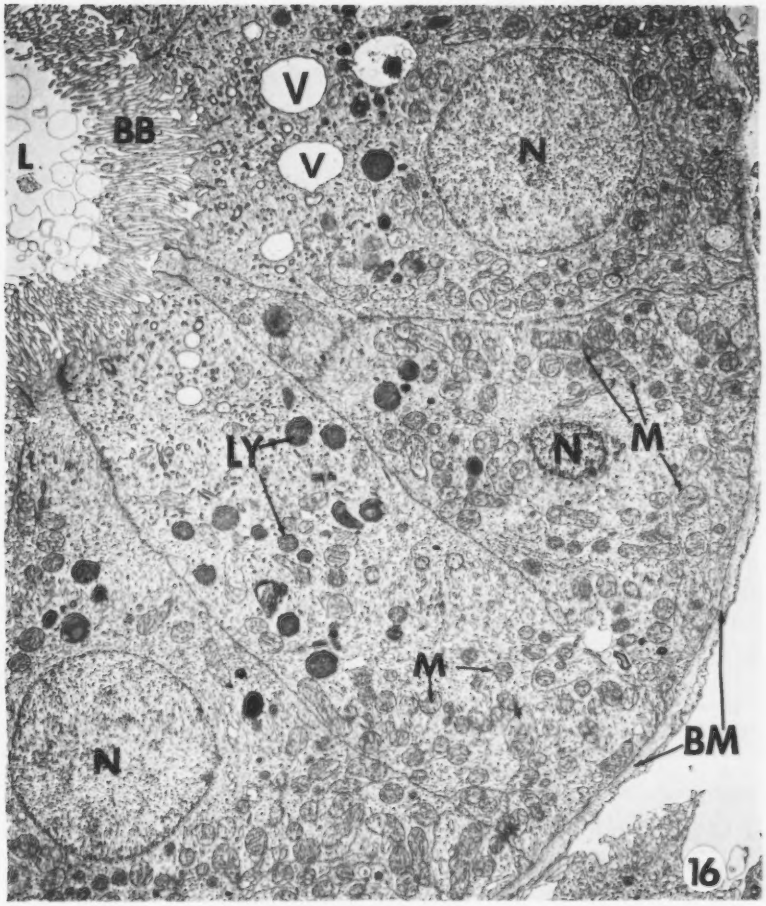


FIG. 16. Electron micrograph of proximal tubule of channel catfish kidney fixed with a combination of glutaraldehyde and paraformaldehyde. ×5500. Jeolco JEM 100B electron microscope. BB, brush border; L, lumen; BM, basement membrane; N, nuclei; V, apical vacuoles; M, mitochondria; LY, lysosomes.

within myocytes due to swollen sarcoplasmic reticulum and mitochondria. The differences in quality of fixation in the heart may be obviated by cutting the aorta, proximal to the site of cannulation, rather than the ventricular wall. This would increase the perfusion pressure within the heart and aid in penetration of fixative.

Although it may not be possible to fix the smallest or largest specimen in this manner, adaptations in size of cannulae, perfusate volume and amount of perfusion pressure should make this technique suitable to a wide variety of fish species. The problems of anoxia, mechanical stress, and inadequate fixative penetration, inherent in most methods of fixation, are largely obviated by whole body perfusion.

Acknowledgments — The work on which this report is based was supported in part by funds provided by the U.S. Department of the Interior, Office of Water Resources Research, as authorized under the Water Resources Research Act of 1964, Contract No. 14-31-0001-3217 and by a grant from the United States Public Health Service for Graduate Training in Pathology, GM-00431-14.

C. R. Pool, University of Louisville, assisted in the early phase of the study and prepared the electron micrograph of the liver. R. T. Jones, University of Maryland, assisted in many of the perfusions and gave editorial assistance.

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Amino Acid Supplementation of Casein in Diets of Atlantic Salmon (*Salmo salar*) Fry and of Soybean Meal for Rainbow Trout (*Salmo gairdneri*) Fingerlings

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RUMSEY, G. L., AND H. G. KETOLA. 1975. Amino acid supplementation of casein in diets of Atlantic salmon (*Salmo salar*) fry and of soybean meal for rainbow trout (*Salmo gairdneri*) fingerlings. J. Fish. Res. Board Can. 32: 422–426.

Fry of Atlantic salmon (*Salmo salar*) fed a diet containing isolated fish protein grew significantly faster than those fed a diet containing casein without supplemental amino acids, and had improved feed conversion and lower mortality. Supplementing the casein diet with essential amino acids to the levels in the isolated fish protein diet significantly increased growth rate, reduced mortality, and improved feed conversion.

When fingerling rainbow trout (*S. gairdneri*) were fed diets containing soybean meal as the sole source of protein, additions of amino acids to simulate those levels of essential amino acids in trout eggs and isolated fish protein significantly improved growth. Additions of methionine, lysine, histidine, and leucine individually and in several combinations had no effect.

Printed in Canada (J3547)
Imprimé au canada (J3547)

RUMSEY, G. L., AND H. G. KETOLA. 1975. Amino acid supplementation of casein in diets of Atlantic salmon (*Salmo salar*) fry and of soybean meal for rainbow trout (*Salmo gairdneri*) fingerlings. J. Fish. Res. Board Can. 32: 422–426

Les alevins de saumon (*Salmo salar*) soumis à un régime contenant une protéine de poisson isolée croissent à un rythme nettement plus rapide que ceux qui ont reçu un régime contenant de la caséine sans amino-acides supplémentaires. Ils ont de plus une meilleure conversion de nourriture et des mortalités plus faibles. Le fait d'ajouter au régime de caséine des amino-acides essentiels aux mêmes concentrations que dans le régime de protéine de poisson accélère notablement la croissance, réduit les mortalités et améliore la conversion de nourriture.

Lorsqu'on donne à des alevins de truite arc-en-ciel (*S. gairdneri*) de la grosseur du doigt des régimes contenant de la farine de soya comme seule source de protéine, l'addition d'amino-acides pour simuler les niveaux d'amino-acides essentiels d'oeufs de truites ou de protéine de poisson isolée améliore la croissance de façon significative. L'addition de méthionine, lysine, histidine et leucine, individuellement ou en plusieurs combinaisons différentes, est sans effet.

Received September 3, 1974
Accepted December 4, 1974

Reçu le 3 septembre 1974
Accepté le 4 décembre 1974

THE objective of this study was to determine whether adding amino acids might improve the nutritional value of casein in diets fed to Atlantic salmon (*Salmo salar*) fry and of conventional dehulled soybean meal in diets fed fingerling rainbow trout (*S. gairdneri*). The experiments were conducted for 14 and 6 wk, respectively. Little is known about the effects of amino acid supplementation of protein sources in the diets of salmonid fishes. Studies at our laboratory and elsewhere (Cowey et al. 1971; Cowey et al. 1974; Nose 1971; Orme and Lemm 1973; Windell et al. 1974) indicate that few proteins from other sources have replaced fish meal protein with reasonable success.

Materials and methods — Atlantic salmon fry (mean weight, 0.5 g; SD, 0.03) were obtained from eggs of sea-run fish captured in the Penobscot River, Maine. The rainbow trout fingerlings (mean weight, 9.0 g; SD, 0.2) were reared from eggs of hatchery stock.

The experimental diets (Table 1) fed to salmon fry were a basal diet containing casein as the source of protein (diet 1), a diet containing isolated fish protein (diet 2), and another diet formulated from basal diet 1 by isonitrogenously substituting several amino acids (footnote d, Table 2) for glutamic acid to raise their levels to those in diet 2.

The rainbow trout fingerlings were fed, by use of demand feeders (Sumari and Westman 1972), a basal diet (diet 3, Table 1) containing soybean meal as a source of protein and nine other diets (Table 3) prepared by adding various amino acids individually and in combinations. Methionine was added (diet 3[1]) to meet the suggested requirement of chinook salmon (*Oncorhynchus tshawytscha*) for total sulfur amino acids (Halver et al. 1959). Histidine and lysine, separately, together, and with methionine (diets 3[2]–3[5]) were added to raise their levels to those in composite whole trout carcasses (*Salmo gairdneri, S. trutta,* and *Salvelinus fontinalis*) (G. L. Rumsey unpublished data). The values for essential amino acids in carcasses used were (in percent of protein): arginine, 5.92; histidine, 3.08; isoleucine, 3.66; leucine, 6.67; lysine, 8.00; methionine, 2.55; methionine + cystine, 3.28; phenylalanine, 3.95; phenylalanine + tyrosine, 7.42; threonine, 3.34; and valine, 4.15. Leucine alone and in combination with either methionine or methionine, lysine, valine, and threonine (diet 3[6]–3[8]) were added to raise their levels to those in whole-egg protein from rainbow trout (Suyama 1958). Methionine, leucine, lysine, valine, threonine, histidine, tryptophan, and tyrosine were all added (diet 3[9]) to raise their levels to those in isolated fish protein (Prot-Animal, Astra Pharmaceutical Products, Inc., Boston, Mass.).[1]

Before the experiment, all fish were acclimated for 2 wk in experimental jars equipped with feeders containing a commercially prepared trout diet. Each diet was fed to triplicate jars of 18 trout each except for the basal diet, which was fed to six jars of trout. Experimental diets were pelleted, broken into crumbles, and sized. The sizes of crumbles fed to fish of various weights were those reported by Rumsey (1972).

For the salmon fry, feeding levels were determined by the method of Buterbaugh and Willoughby (1967) using a hatchery constant of 7.0. The fry were reared in duplicate 150-liter tanks each containing 300 fish and supplied with heated (13.5–14.5 C), aerated well water at 7.5 liters/min. The trout fingerlings were reared in 7-liter jars supplied with spring-fed pond water (temperature 8–17 C) at about 3 liters/min.

The duration of the experiment with the trout was considered sufficient because of the similarity in results at 6 and 14 wk in the experiment with salmon and on the basis of published works with rainbow trout and other fish showing experiments of 2–6 wk to be adequate in the study of proteins or amino acids or both (Brett 1971; Nose 1971; Gerking 1971; DuPree and Halver 1970; DeLong et al. 1959; Ogino and Saito 1970).

[1]Reference to trade names does not imply Government endorsement of commercial products.

TABLE 1. Percentage compositions of the three diets tested.

Ingredient	Diet 1	Diet 2	Diet 3
Casein	40.0	–	–
Isolated fish protein[a]	–	40.0	–
Soybean meal[b], 49% protein	–	–	80.0
Dextrin, tapioca	20.0	20.0	6.5
L-glutamic acid	6.2	5.2	–
Cellulose	21.3	21.3	–
Constant[c]	13.5	13.5	13.5

[a]Prot-Animal, Astra Pharmaceutical Products, Inc., Boston, Mass.

[b]Finely ground.

[c]In percent of diet: herring oil, 10; carboxymethylcellulose, 1.0. Vitamins in mg/kg of diet: thiamine HCl, 50; riboflavin, 200; pyridoxine HCl, 50; niacin, 750; D-Ca-pentothenate, 500; inositol 2000; D-biotin, 5; folic acid, 15; L-ascorbic acid, 1000; cyanocobalamine, 0.1; menadione sodium bisulfite, 40; choline Cl, 5000. Vitamins in IU/kg of diet: DL-alpha-tocopherol acetate, 500; vitamin A palmitate, 10,000; vitamin D_3, 4000. Minerals in mg/kg of diet: $CaCO_3$, 250; KCl, 4672; KH_2PO_4, 4000; Na_2HPO_4, 3087; $MgSO_4$, 2475; $FeSO_4 \cdot 7H_2O$, 250; $ZnSO_4 \cdot 7H_2O$, 220; $MnSO_4 \cdot H_2O$, 92; $CuSO_4$, 20; KI, 1.3; $(NH_4)_6Mo_7O_{24} \cdot 4H_2O$, 0.4; $CoCl_2 \cdot 6H_2O$, 0.2; Na_2SeO_3, 0.2; $CrCl_3 \cdot 6H_2O$, 0.5. Diet 3, NaCl, 0.5%.

Experimental data were subjected to analysis of variance and Duncan's multiple range test (Steel and Torrie 1960).

For the salmon, we determined body composition using duplicate group samples of 20 fry each at the beginning and end of the experiment. Fish were fasted 24 h prior to sampling. Samples were macerated in a blender with dry ice to a homogeneous slurry. Moisture and ash contents were determined immediately; the rest of the sample was lyophilized and later analyzed for fat and protein according to A.O.A.C. methods (Horowitz 1970). We calculated protein efficiency using the expression:

[(carcass protein at 14 wk)–(carcass protein at beginning)] × (100) ÷ (protein fed).

At the end of the experiment, 10 fish from each treatment were examined by a veterinary pathologist for gross pathological and histopathological changes.

Results — Salmon fry fed the diet containing isolated fish protein grew significantly ($P < 0.05$) faster than those fed diets containing casein without supplemental amino acids and had the lower feed:gain ratio and mortality (Table 2). Supplementation of the casein diet with amino acids significantly increased growth rate, improved feed conversion (feed:gain), and reduced mortality. Results at the end of 6 wk were similar.

The mortality when salmon fry were fed the diet containing casein without supplemental amino acids was especially high between 6 and 8 wk. Examination of the dead fish revealed an epithelial hyperplasia of the gill lamellae and the presence of myxobacteria associated with bacterial gill disease. At the end of the experiment, however, no gross pathological or remarkable histopathological changes were found in any of the surviving salmon.

The apparent efficiency of deposition of dietary protein into body tissue (Table 2) was increased nearly twofold by amino acid supplementation of the casein diet. Apparent efficiency for the casein diet with supplements was identical to that for the diet containing isolated fish protein.

Salmon fed the casein diet without supplements tended to have higher carcass moisture, ash, and protein contents and lower fat content than those fed isolated fish protein. Supplementation with amino acids prevented these changes.

Although growth of the trout (Table 3) was slow, adding certain combinations of amino acids to the soybean meal diet (diets 3[8] and 3[9]) significantly improved growth. The combination of methionine, leucine, lysine, valine, threonine, histidine, tryptophan, and tyrosine, as well as the combination of methionine, leucine, lysine, valine, and threonine significantly increased growth rate. In contrast, supplements of methionine, histidine, lysine, and leucine, alone and in various combinations, had no significant effect on growth. Mortality did not exceed 6% for any treatment.

Unpublished results (H. G. Ketola 1973) with rainbow trout showed that supplementation of a soybean meal diet with graded levels of DL-methionine up to 0.8% of diet had no significant effect on growth.

Discussion — The significant improvement in growth when casein and soybean meal were supplemented with mixtures of amino acids to simulate fish protein suggests a potential for improvement in protein utilization by salmonids when fed sources of protein other than fish. The marked improvement in utilization of protein when casein was supplemented with amino acids confirms similar observations with poultry (Dean and Scott 1965; Solberg 1971; Velu et al. 1971).

The mortality apparently associated with bacterial gill infection, solely in fish fed the diet deficient in amino acids, suggests a causal relation between development of the disease and the

TABLE 2. Growth characteristics and mortalities of Atlantic salmon (*Salmo salar*) fry (duplicate lots of 300 fish each) fed a casein diet, with and without supplemental amino acids, and a semipurified source of fish protein.

Diet	Wt gain[a] (g)	Feed: gain (g:g)	Mortality (%)	Carcass composition				Protein efficiency[b] (%)
				H_2O (%)	Ash (% of dry matter)	Fat (% of dry matter)	Protein (% of dry matter)	
Casein (diet 1)	1.2A[c]	3.9A	61	77	10	20	67	13
+amino acids[d]	2.6B	1.9B	8	74	8	27	61	24
Isolated fish protein (diet 2)	3.1C	1.8B	5	74	9	26	60	24
Pooled SE	0.04	0.2						

[a]Average 14-wk weight gain of salmon having an initial body weight of 0.5 g. Average gains at 6 wk for the three respective diets were 0.2A, 0.5B, and 0.7C (pooled SE = 0.03).

[b]Apparent efficiency of deposition of dietary protein into tissue [(protein deposited ÷ protein fed) × 100].

[c]Values not followed by the same letter are significantly different ($P < 0.05$).

[d]The following amino acids were substituted for glutamic acid (% of diet): L-arginine, 0.97; L-cystine, 0.29; L-lysine, 0.64; L-methionine, 0.14; L-threonine, 0.18; and L-tryptophan, 0.16.

TABLE 3. Gains in weight for fingerling rainbow trout (*Salmo gairdneri*) (triplicate lots of 18 fish each) fed for 6 wk a diet containing soybean meal (diet 3) with and without supplemental essential amino acids.

Diet	Supplement	Average gain[a] (g)
3	None	4.2A[b]
3[1]	Met[c] (0.3)[d]	4.6AB
3[2]	His (0.3)	4.2A
3[3]	Lys (0.7)	3.4A
3[4]	His+Lys	4.5AB
3[5]	His+Lys+Met	4.4AB
3[6]	Leu (0.7)	4.4AB
3[7]	Met+Leu	4.8AB
3[8]	[Met+Leu+Lys+Val+Thr][e]	6.0BC
3[9]	[Met+Leu+Lys+Val+Thr+His+Trp+Tyr][f]	6.4C
	Pooled S.E.	0.3

[a]Average 6-wk gain of trout having an initial body weight of 9 g.

[b]Values not followed by the same letter are significantly different ($P < 0.05$).

[c]Met, methionine; his, histidine; lys, lysine; leu, leucine; val, valine; thr, threonine; trp, tryptophan; tyr, tyrosine.

[d]Values in parentheses are levels of supplementation in percent of diet.

[e]Percentages of supplements in diet were 0.3, 0.7, 0.4, 0.3, and 0.3, respectively.

[f]Percentages of supplements in diet were 0.5, 0.3, 1.4, 0.5, 0.6, 0.1, 0.3, and 0.1, respectively.

deficiency. For terrestrial animals, causal relations between deficiencies of protein, amino acid, and minerals in diets and predisposition to bacterial, viral, and parasitic infections have been reported (Hill 1974; Scrimshaw et al. 1968). Although causal relations between amino acid deficiencies and infectious diseases have not been reported in fish, Snieszko (1972) summarized works showing that dietary vitamin deficiencies predisposed Atlantic salmon to protozoan infections.

Slow growth of rainbow trout fingerlings when fed soybean meal was also observed by Nose (1971), even though digestion of the protein by rainbow trout in his experiments was only slightly reduced. Some investigators, however, showed that soybean meal was very poorly digested by young rainbow trout and plaice (Windell et al. 1974; Cowey et al. 1974). Andrews and Page (1974) observed that poor growth of young catfish fed diets containing soybean meal was not improved by dietary supplements of methionine, cystine, or lysine.

Shanks et al. (1962) showed that the qualitative requirements of rainbow trout were similar to those of chinook and sockeye salmon (Halver et al. 1957; Halver et al. 1968; Halver et al. 1959; Halver and Shanks 1960; DeLong et al. 1962; Chance et al. 1964; Halver 1965; Klein and Halver 1970). The results of the experiment with fingerling trout suggest that the quantitative re-

quirements of fingerling rainbow trout are approximated by the amino acid patterns in isolated fish protein and the protein in eggs of rainbow trout or, alternatively, that soybean meal contains disproportionate levels of amino acids causing a reduction in growth rate which is corrected by additions of specific amino acids (Harper et al. 1970).

Acknowledgments — For his careful examination of experimental salmon we thank Dr J. M. King, Department of Veterinary Pathology, College of Veterinary Medicine, Cornell University, Ithaca, N.Y.

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PERSPECTIVES

Dynamics of Exploited Whitefish Populations and their Management with Special Reference to the Northwest Territories

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HEALEY, M. C. 1975. Dynamics of exploited whitefish populations and their management with special reference to the Northwest Territories. J. Fish. Res. Board Can. 32: 427–448.

Available data on mortality, growth, reproduction, and stock size in exploited and unexploited populations of lake whitefish (*Coregonus clupeaformis*) are reviewed with a view to understanding the dynamics of exploited populations and improving their management. Natural mortality ranged from about 0.20 to 0.80 in unexploited populations. In exploited populations total mortality was as high as 0.94. Unexploited populations showed a wide range of growth rates. Growth rate increased with increasing exploitation, and growth rate in all heavily exploited populations was similar to the most rapid growth rate shown by unexploited stocks. Heavily exploited whitefish matured at a younger age and possibly also at a smaller size than those which were unexploited. Limited data on stock size suggest that although total population size declines under heavy exploitation, the vulnerable population remains of similar size.

It is concluded that whitefish respond to fluctuations in population size through compensatory changes in growth rate. The difference between growth rate in a population and maximum growth rate is a measure of its scope for compensating for increased mortality. Populations with slow growth rate and low mortality should, therefore, have the best fishery potential, while those with high growth rate and high mortality have a low fishery potential. Further, it is possible to judge the fishery potential of a population or its stage of exploitation from relatively simple measurements of mortality, growth, age structure, and maturity.

HEALEY, M. C. 1975. Dynamics of exploited whitefish populations and their management with special reference to the Northwest Territories. J. Fish. Res. Board Can. 32: 427–448.

Afin de mieux comprendre la dynamique des populations exploitées et d'en améliorer la gestion, nous faisons la revue des données disponibles sur la mortalité, la croissance, la reproduction et l'étendue des stocks de populations exploitées et inexploitées de grands corégones (*Coregonus clupeaformis*). La mortalité naturelle est comprise entre 0.20 et 0.80 environ chez les populations inexploitées. Chez les populations exploitées, la mortalité totale peut s'élever à 0.94. Les populations inexploitées montrent une gamme étendue de rythmes de croissance. Le rythme de croissance augmente en fonction d'une exploitation accrue, et le rythme de croissance de toutes les populations fortement exploitées est semblable au rythme maximal observé chez les stocks inexploités. Les grands corégones soumis à une exploitation intense atteignent la maturité sexuelle plus tôt et possiblement aussi à une taille plus petite que ceux qui sont inexploités. Des données limitées sur l'étendue des stocks suggèrent qu'en dépit du fait que la taille globale de la population diminue lorsque cette dernière est fortement exploitée, la population vulnérable demeure de la même grandeur.

Nous en concluons que la grand corégone réagit aux fluctuations de grandeur de population par des changements compensateurs du rythme de croissance. La différence entre le rythme de croissance au sein d'une population et le rythme de croissance maximal donne une mesure de sa

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Printed in Canada (J3403)
Imprimé au Canada (J3403)

faculté de compensation face à des mortalités accrues. Les populations à rythme de croissance lent et à faible mortalité devraient donc offrir le meilleur potentiel de pêche, alors que les populations à rythme de croissance rapide et à forte mortalité ont un faible potentiel. De plus, il est possible de juger du potentiel de pêche d'une population ou de son stade d'exploitation à partir de mesures simples de mortalité, croissance, groupes d'âge et maturité.

Received April 29, 1974
Accepted December 9, 1974

Reçu le 29 avril 1974
Accepté le 9 décembre 1974

WHITEFISH in Canada's lakes are an abundant and valuable resource. On the arctic and subarctic mainland whitefish may be the only species that offer a reasonable fishery harvest in lakes. Of the several species involved, the lake whitefish (*Coregonus clupeaformis*) is currently the most valuable. With the exception of Great Slave Lake, lakes in the Northwest Territories are harvested according to the rule that ½ lb/acre per year of whitefish and trout is the maximum allowable harvest. Although management personnel recognize that this rule is unsatisfactory, a satisfactory alternative has not been put forward. In the provinces, management is more tailored to the characteristics of the individual lakes. Yet fisheries still fail.

In the Northwest Territories, Great Slave Lake is the only lake that has been continuously exploited with success. The fishery is a gillnet fishery and minimum mesh size is 5½-inch (140-mm) stretched mesh. During the early 1960s the harvest appeared to have stabilized at about 3.8 million lb (1.7 million kg) of whitefish annually. Since 1965, catches have declined, however, and in 1973 only 2.2 million lb (1 million kg) were captured (Bond and Turnbull 1973; W. A. Bond personal communication). This recent decline in catches necessitates a reevaluation of the present management of the lake. Most important is the fact that fishery statistics collected from the lake did not permit a decline to be predicted. This is also true of other instances of declines in whitefish fisheries (e.g. Davidoff *et al.* 1973).

There is now considerable background information on the biology of whitefish. The purpose of this paper is to review our knowledge of the biology of lake whitefish and their response to exploitation, and to propose ways to get the best yield from whitefish populations. We need to know what measurements are necessary to judge the fishery potential of virgin populations and to assess the state of exploited populations. I shall examine four parameters relevant to production and yield in whitefish populations: mortality, growth, population size, and reproduction. I shall discuss estimates of these parameters and their variability in both exploited and unexploited populations. Differences between exploited and unexploited populations provide clues to the dynamics of exploited populations and these differences will be a basis for statements about management.

Mortality

Comparison of fishing and natural mortality rates provides insight into the impact of the fishery on the population. This comparison may be approached in two ways, through calculation of fishing and natural mortality rates in exploited populations, and through comparison of total mortality in both exploited and unexploited populations. Further, comparison of mortality rates among unexploited populations contributes to the understanding of natural variability in population parameters and permits better interpretation of data from exploited populations.

UNEXPLOITED POPULATIONS

All published estimates of mortality for unexploited populations are from age structure data (Ricker 1947; Kennedy 1963). I was able to calculate additional mortality estimates from age structure data of other authors (Edsall 1960; Wong and Whillans 1973; Kennedy 1949; M. C. Healey unpublished data). When calculating mortality rate I used techniques described by Ricker (1958). Natural mortality rates for 13 unexploited populations, estimated from age structure data, ranged from 0.18 to 0.74 (Table 1).

EXPLOITED POPULATIONS

Mortality estimates for exploited populations come from both mark-recapture and from age structure data. Mark-recapture estimates of mortality are available for different areas of Lake Huron (Cucin and Regier 1965; Spangler 1970), and tag return data from which mortality can be calculated are available for Lake Huron (Budd 1956) and Lake Winnipeg (Kennedy 1954). Mortality rates estimated from age structure data are published for five lakes (Great Slave, Kennedy 1953, Bond and Turnbull 1973; Winnipeg,

TABLE 1. Mortality estimates for exploited and unexploited whitefish (*Coregonus clupeaformis*) populations.

Lake	Reference	Estimating procedure	Natural mortality		Total mortality	
			Range	Mean	range	mean
Georgian Bay	Budd 1956	Tag returns				0.77
Georgian Bay	Cucin and Regier 1965	Mark recapture	0.23–0.50	0.34	? 0.80	?
Georgian Bay	Cucin and Regier 1965	Catch curve			0.48–0.95	0.62
Huron	Spangler 1970	Mark recapture	0.75–0.98	0.86	0.76–0.99	0.89
Huron	Budd and Cucin 1962	Catch curve				0.93
Huron	Budd 1956	Tag returns			0.53–0.59	0.56
Winnipeg	Kennedy 1954	Tag returns				0.40
Winnipeg	Kennedy 1954	Catch curve			0.50–0.77	0.64
Winnipeg	Davidoff et al. 1973	Catch curve		0.51	0.50–0.96	0.79
Ontario	Christie 1963	Catch curve			0.38–0.69	0.52
Pigeon	Miller 1947	Catch curve			0.50–0.82	0.75
Pigeon	Bidgood 1972	Catch curve				0.66
Wabamun	Miller 1956	Catch curve			0.50–0.83	0.66
Shakespeare	Ricker 1947	Catch curve	0.08–0.45	0.18		
Opeongo	"	Catch curve	0.41–0.59	0.48		
Opeongo Dwarf	"	Catch curve	0.64–0.78	0.72		
Nipigon	"	Catch curve				0.36
Michigan	Roelofs 1958	Catch curve			0.84–0.98	0.94
La Martre	Bond 1973	Catch curve				0.65
Hottah	Wong and Whillans 1973	Catch curve		0.46		
Nueltin	Kennedy 1963	Catch curve		0.57		
MacDonald	"	Catch curve		0.71		
MacEwan	"	Catch curve		0.74		
Great Bear	Kennedy 1949	Catch curve		0.42		
Great Slave	Kennedy 1953	Catch curve			0.51–0.63	0.57
Great Slave	Bond and Turnbull 1973	Catch curve				0.58
Red lakes	Peterka and Smith 1970	Catch curve			0.39–0.91	0.75
Red lakes	Peterka and Smith 1970	Catch-per-unit-effort			0.20–0.71	0.48
Munising Bay	Edsall 1960	Gillnet catch		0.44		
Munising Bay	Edsall 1960	Trawl catch		0.26		
Erie	Van Oosten and Hile 1947	Trapnet catch			0.68–0.81	0.74
Erie	Van Oosten and Hile 1947	Gillnet catch			0.67–0.74	0.71
Erie	Lawler 1965	Catch curve			0.40–0.94	0.74
Alexie	Healey (unpublished data)	Catch curve		0.48		
Baptiste		Catch curve		0.56		
Chitty		Catch curve		0.54		
Drygeese		Catch curve		0.56		
La Ronge	Qadri 1968	Catch curve				0.50
Buck	Bidgood 1972	Catch curve				0.59

Kennedy 1954, Davidoff et al. 1973; Michigan, Roelofs 1958; Red lakes, Peterka and Smith 1970; and Lac la Martre, Bond 1973). Age structure data from which I calculated mortality are published for seven other lakes (Erie, Lawler 1965, Van Oosten and Hile 1947; Ontario, Christie 1963; Huron, Cucin and Regier 1965; Pigeon, Miller 1947, Bidgood 1972; Wabamun, Miller 1956; Buck, Bidgood 1972; La Ronge, Qadri 1968).

Estimates of total mortality for 14 exploited populations ranged from 0.36 to 0.94. Estimates for six of the populations were greater than 0.74, the highest rate observed in unexploited populations (Table 1).

Cucin and Regier (1965) and Spangler (1970) estimated both natural and total mortality from their mark-recapture data, while Davidoff et al. (1973) were able to estimate natural mortality from the regression of instantaneous total mortality on effect. Cucin and Regier's estimate of natural mortality for Georgian Bay whitefish (0.34) and

Davidoff, Rybicki and Doan's estimate for Lake Winnipeg (0.51) are within the range for unexploited populations, while Spangler's estimate for the south Manitoulin Island (Lake Huron) population (0.86) was higher than the highest value for unexploited populations (Table 1). The south Manitoulin Island population was suffering heavy lamprey predation, however.

Total mortality in exploited population is, on average, higher than mortality in unfished stocks. Unfortunately, the data are not sufficient in most instances to separate natural and fishing mortality. Cucin and Regier (1965) estimated that fishing mortality in Georgian Bay ranged from 70 to 80%. Spangler (1970) estimated that fishing mortality at south Manitoulin Island was 7% for 2-year-olds, 47% for 3-year-olds, and 60% for 4-year-olds. In Lake Winnipeg natural mortality was 0.51 and total mortality ranged from 0.50 to 0.93 in the period 1944–69 (Davidoff et al. 1973). Fishing mortality, therefore, ranged from near 0.0 to 86% (Ricker 1958, equation 1.7).

These fishing mortality rates for populations suffering moderate to heavy exploitation may be compared with the overall picture for exploited whitefish (Table 1). Average mortality of unexploited populations is 0.49 and of exploited populations is 0.64 (geometric means of mortality rates for individual populations in Table 1). Average fishing mortality is therefore 0.29. Considering that for the more heavily fished populations in Table 1 fishing mortality is probably in the range 0.5–0.7, the fishing mortality of the lightly fished populations must be low to produce an average fishing mortality of only 0.29.

Sources of Bias in Mortality Estimates

The wide variation in mortality rates among both unexploited and exploited populations may be of considerable significance in management. First, however, I must consider how much of this variation is real and how much is a result of biases in the data.

In calculating mortality rates from their data both Spangler (1970) and Cucin and Regier (1965) assumed that the requirements for a Petersen type estimate of population size were not seriously violated. Many of the possible sources of error in mortality estimates from mark-recapture data (tag loss, failure to report recaptures, differential mortality of tagged fish) would cause mortality to be overestimated, however. If these estimates are biased, therefore, they are more likely to be overestimates than underestimates.

In calculating mortality from Budd's (1956) recapture data for south Manitoulin Island, I assumed that there was no tag loss and that no fish captured outside South Bay were returned to the population. Both these assumptions are probably incorrect, but they bias the mortality estimate in opposite directions. In Georgian Bay Budd (1956) had more recaptures the year after tagging than the year of tagging. The fish for tagging were captured in pound nets, whereas recaptures were mainly from gillnets. Cucin and Regier (1965) concluded that poundnet-captured fish were too small to be fully vulnerable to gillnets until the following year. I, therefore, assumed that Budd's marked fish in Georgian Bay were not fully vulnerable to the recapture gear until the year after marking and calculated mortality accordingly. In calculating mortality from Kennedy's (1954) tag returns for Lake Winnipeg, I first corrected the number of returns for intensity of fishing in two ways: according to the number of licensed vessels and according to total catch of fish. Both corrections result in the same mortality estimate (0.40). As with Cucin and Regier's (1965) and Spangler's (1970) estimates, these estimates are more likely to be too high than too low. On average, therefore, mark-recapture estimates of mortality may be too high, yet the observed range of mortality estimates is still very wide (0.40–0.99, Table 1).

The age structure data from which mortality was estimated were mainly from gillnet sampling, although some data were from trawl and trapnet sampling. All these data suffer from net selection of one form or another, and must be interpreted accordingly (McCombe and Fry 1960; Regier and Robson 1966). Both the youngest and oldest fish are relatively less susceptible to capture than intermediate-sized fish, so that mortality estimates from catch curves are generally overestimates. This bias is the more serious, the greater the growth of fish in the age ranges considered, and the more restricted the range of mesh sizes used. In the studies reported here, the range of mesh sizes used was generally adequate to sample all the larger size-classes of whitefish. The extent of bias will depend mainly, therefore, on the growth of the older fish.

For the three unexploited populations examined by Ricker (1947), mortality rates cover almost the whole observed range (0.18–0.72, Table 1). Whitefish from Shakespeare Island Lake have a very slow growth rate and live to great age (27 years). The total population's size is small so that one must suppose that mortality rate is also low, as was found by Ricker (0.18). Estimates of mortality for the normal and dwarf races of white-

fish in Lake Opeongo (Kennedy 1943) were quite different (Ricker 1947). The normal fish almost doubled in size over the ages for which mortality was calculated, so this estimate (0.43) may be too high. Ricker calculated mortality age-class by age-class, however, so the bias due to growth should be minimal. Ricker considered his mortality estimate for the dwarf race (0.72) to be too low. Since small mesh nets do not adequately sample small fish of the normal race, the estimate for the dwarf race may be too low. Considerable growth over the ages involved in mortality estimation may have biased estimates for Hottah Lake (Wong and Whillans 1973), Munising Bay, Lake Superior (Edsall 1960), and McEwan Lake (Kennedy 1963). This source of bias was probably less important in estimates for Great Bear Lake (Kennedy 1949), Nueltin and MacDonald lakes (Kennedy 1963), and Alexie, Baptiste, Chitty, and Drygeese lakes (M. C. Healey unpublished data). All of the fish from the exploited populations were growing rapidly over the ages for which mortality was estimated, so that this source of bias is likely to be more pronounced in the estimates for exploited populations than in those for unexploited populations.

A further source of bias in mortality estimates from age structure data is from incorrect aging. Aging of whitefish is normally done from scale annuli. Scales from very old fish are sometimes difficult to read and fish may be underaged, thus causing mortality estimates to be too high. Underaging is less likely to be a problem in heavily exploited stocks where there are few old fish.

Although it is possible to identify these sources of bias, it is usually not possible to estimate the amount of bias. A qualitative estimate of the bias in mortality rates may be gained by comparing estimates from two or more methods or data sets. Several studies permitted such a comparison. Peterka and Smith (1970) calculated the survival rate of each year-class in the population in Red lake, Minnesota from catch-per-unit-effort figures. Mortality for each age-class ranged from 0.20 to 0.71 with a mean of 0.48. Mortality calculated from age structure data ranged from 0.39 to 0.91 with a mean of 0.75. Mortality estimated from trawl samples (0.26) was considerably less than mortality estimated from gillnet samples (0.44) in Munising Bay, Lake Superior (Edsall 1960). Mortality estimates based on the age structure of fish captured in trapnets (0.71) and gillnets (0.74) in Lake Erie between 1927 and 1930 were similar and similar to the estimate from combined gillnet and pound net catches reported by Lawler (1965) for later years (1929–56) (Table 1). Kennedy (1954) calculated the mortality of whitefish in Lake Winnipeg from age structure data to be 0.64, substantially more than the mortality (0.40) suggested by tag returns. The highest values of total mortality from Georgian Bay calculated by mark-recapture methods (Cucin and Regier 1965) (0.77–0.80) are less than the highest values calculated from age structure data (0.90 and 0.95 for 1963 and 1964), although the average mortality over all years of age structure data (0.62) seems reasonable and is less than the estimate made from Budd's (1956) earlier tag return data (0.77). The estimates of total mortality reported by Cucin and Regier refer to a particular year-class for a particular year of life. These may be compared with catch curve estimates for the same year-classes and ages. Corresponding mortalities are 0.80, 0.80; 0.77, 0.71; 0.78, 0.50 for mark-recapture and gillnet catch curve estimates, respectively.

Two additional sources of variability in the estimates of mortality are changing mortality with age and variable year-class strength. Mortality rates were calculated over different spans of age depending on the shape of the catch curve which in turn depended on the type of gear used. Older fish appear to have a somewhat higher mortality rate than intermediate-aged fish, so that mortality estimates will be higher when based only on older aged fish than when based on a wider range of ages. The amount of variation introduced by changing mortality with age is difficult to judge because there is so little good information on changes in mortality with age. However, in most instances estimates of mortality were based on a wide enough range of ages that the contribution to variability in the estimates from this source will be small.

In some populations, especially those which are heavily fished, strong and weak year-classes are evident (Lawler 1965; also data in Cucin and Regier 1965; Christie 1963; Kennedy 1954). Mortality of individual year-classes is different but there is no consistent relationship between mortality and year-class strength (Table 2). If only 1 or 2 years worth of data are available, the presence of a strong year-class which was becoming fully vulnerable would make mortality calculated from the age structure higher than the true mortality. The converse would be true if a weak year-class were becoming vulnerable at the time of sampling. This source of variation does not contradict the conclusion that mortality is extremely variable between populations of whitefish for two reasons. In uncropped populations with a relatively old age structure the mechanism of damping out year-class variation appears to be quite effective so that large oscillations do not

TABLE 2. Mortality of individual year-classes in four lakes in relation to the size of their contribution to the fishery. Contribution to the fishery is ranked largest to smallest.

	Lake and source				
	Erie	Huron	Ontario	Winnipeg	
Ranked contribution	Lawler (1965)	Cucin and Regier (1965)	Christie (1963)	Kennedy (1954)	Davidoff et al. (1973)
1	0.68	0.53	0.54	0.58	0.62
2	0.66	0.72	0.69	0.52	0.70
3	0.67	0.68	0.48	0.69	0.84
4	0.88	0.80	0.53	0.57	0.89
5	0.56	0.59	0.67	0.66	0.88
6	0.73	0.80	0.55	0.64	0.79
7	0.83	0.74	0.57	0.54	0.77
8	0.68	0.68	0.31	0.47	0.51
9	0.86		0.60	0.67	0.64
10	0.69		0.55		0.90
11	0.65		0.45		0.63
12	0.71				0.54
13					0.76

occur. Further, a comparison of survival of individual year-classes for those populations where sufficient records have been kept indicates that survival can vary as much between year-classes as it can between years. This means that, although 1 or 2 years of sampling may not produce a mortality estimate that is characteristic of the population, it is unlikely to produce an estimate that is outside the range of values for the population.

CONCLUSIONS

Although some biases and inconsistencies are evident in the estimates of mortality rate, some conclusions are possible. The estimates of mortality are consistent with such other factors as population size, longevity, and intensity of fishing. In general, the biases considered would result in overestimates of mortality rate rather than introducing a large random error. Where comparisons could be made, estimates of mortality from gillnet catch curves were the same as or greater than estimates by other methods, suggesting that the bias in estimates from catch curves may be higher than in estimates by other methods. Random variation in the estimates could result from both increasing mortality with age and variations in year-class strength. Nevertheless, I consider that the range of mortality rates calculated for unexploited whitefish populations is appropriate and suggests a very wide range of natural mortality coefficients. Further, the comparison of total mortality rate in both exploited and unexploited populations indicates that, for many populations, fishing mortality contributes only a small amount to total mortality.

Growth Rates

Rate of growth is a fundamental variable in calculations of production; yet, I know of no published comparisons of growth among exploited and unexploited whitefish populations. Considerable information exists, however, on whitefish growth. I have not tried to collect every bit of information on growth. In particular I have not delved too deeply into manuscript reports of provincial government agencies, although I have made use of some of these data. My purpose is to examine growth in exploited and unexploited populations and to emphasize differences which may reflect responses to exploitation.

I shall present all data on growth as fork length in millimeters at annular age. When data were not given in this form I made the appropriate conversions. When growth in weight was reported together with data on length and weight, I used these data to convert weight at age to length at age. Where length–weight data were not given I used an average formulation from a range of data:

$$W_{(g)} = 0.004L_{(cm)}^{3.3}$$

The length exponent in the length–weight relationship for whitefish varies from about 3.1 to 3.5, so use of an average formula may introduce some bias. This calculation was never applied to any extremes of growth, however.

When total or standard length was reported I converted to fork length by the following formulae:

$$L_{(fork)} = \frac{L_{(tot)} + 0.1456}{1.12}$$

$$L_{(fork)} = \frac{L_{(std)} - 0.776}{0.889}$$

These conversion relationships come from data I collected on a number of populations in the Northwest Territories.

Data on growth come from a wide variety of geographic locations and lake types scattered throughout the zoogeographic range of whitefish (Fig. 1).

Unexploited Populations

Unexploited populations showed considerable variability in rate of growth (Table 3, Fig. 2A). The slowest growing populations occurred in Trout Lake, Wisconsin (Hile and Deason 1934) and in Munising Bay, Lake Superior (Edsall 1960). Most rapidly growing populations were from McEwan Lake (Kennedy 1963) and Kam Lake (Healey and Woodall 1973) in the Northwest Territories. Fish from the slowest growing populations grew roughly half as fast as those from the fastest growing populations.

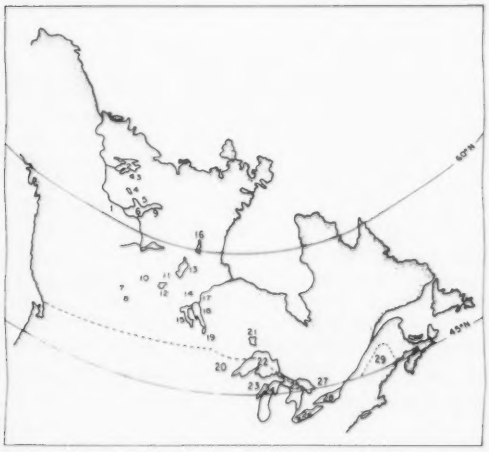

FIG. 1. Locations of lakes referred to in this manuscript: 1. MacEwan, 2. Great Bear, 3. Hottah, 4. La Martre, 5. Alexie, Baptiste, Chitty, Drygeese, Long, Kam, Grace, Michel, Dunnet, Trout, 6. Great Slave, 7. Wabamun, 8. Pigeon, Buck, 9. MacDonald, 10. Big Peter Pond, 11. MacKay, Hebden, Contact, Sulphide, 12. La Ronge, 13. Reindeer, 14. Morton, File, Big Sand, Williams, 15. Neso, Payuk, Mink Narrows, Big Athapapuskow, Little Athapapuskow, Big Twin, Little Twin, 16. Nueltin, 17. Landing, 18. Winnipeg, 19. George, 20. Red lakes, 21. Nipigon, 22. Superior, 23. Trout, 24. Michigan, 25. Huron, 26. Erie, 27. Opeongo, Shakespeare Island, 28. Ontario, 29. Cliff, Second Musquacook, Webster.

Current management practice in the north is based on the supposition that growth in northern fish populations is slow and that their productivity is, therefore, low. An examination of the facts, however, indicates the opposite. Growth curves for fish from the Northwest Territories cluster in the upper portion of the observed range of growth rates for lake whitefish (Fig. 2B). Whitefish north of 60°N do not grow slowly; on the contrary they grow quickly. Kennedy (1949) also made this observation in discussing the growth of Great Bear Lake whitefish. To emphasize the point, let us examine the growth of whitefish from Great Bear and Nueltin lakes, perhaps the coldest and most oligotrophic lakes for which data are available. Growth curves for these lakes are those numbered 11 and 5 in Fig. 2B. Fish from Nueltin Lake have an intermediate growth rate, whereas those from Great Bear grow at a rate above average for the Northwest Territories.

Growth in the 32 unexploited populations may further be examined in relation to the climate of the area where they occur. Keeping in mind that climate is only an indicator of lake temperatures, I divided the whitefish populations for which I have growth data into three approximately equal groups according to the number of degree days above 42 F (5.5 C) in the region where they occur. These three groups were those living where degree days were: fewer than 1600 (890 C), between 1600 and 2200 (890–1220 C), and more than 2200 (1220 C). Poorest growth was shown by populations living where degree days exceeded 2200 (1220 C) (Fig. 2C). Best growth was in the range 1600–2200 (890–1220 C) degree days, although growth of populations where degree days were fewer than 1600 (890 C) was hardly lower than the best growth.

It can be concluded, therefore, that the lake whitefish is a cold-adapted species, growing best in the central and northern parts of its range and poorest in the southern, warmer parts of its range. Therefore, if production is low in northern populations it is not due to poor growth of the whitefish.

Sources of Variation

Although growth and climate appear related, it is not possible to make precise predictions about

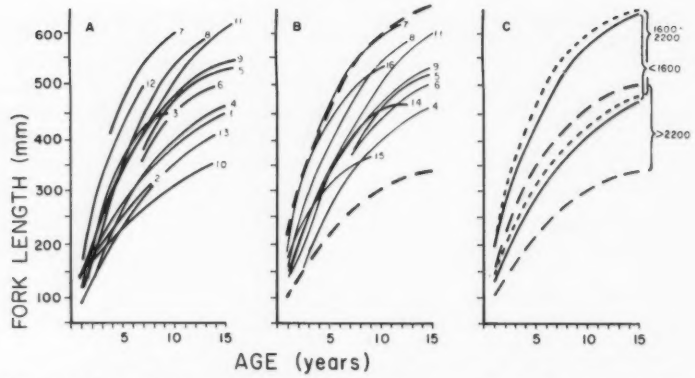

FIG. 2. Growth in unexploited whitefish (*Coregonus clupeaformis*) populations. A, Representative growth curves for populations from the whole zoogeographic range of whitefish. B, Growth in Northwest Territories populations compared with total range in growth from A (dashed lines). C, Range of growth for populations in different climatic zones (<1600 degree days above 42 F; 1600–2200 degree days; >2200 degree days). Lakes are: 1. Shakespeare Island, 2. Opeongo, 3. George, 4. Great Slave, 5. Nueltin, 6. MacDonald, 7. MacEwan, 8. La Martre, 9. Hottah, 10. Munising Bay, 11. Great Bear, 13. Big Athapapuskow, 13. Trout (Wisconsin), 14. Alexie, 15. Long, 16. Kam.

growth from temperature data, for within any climatic zone growth still varies widely. There is not sufficient data on most lakes to further explore the sources of this variation. However, three sources of variation which are difficult to eliminate from most growth data are worth mentioning. The first is the seasonal growth pattern of the fish. Samples for estimating growth rate were taken at different times of year. Whitefish grow mainly during summer (Kennedy 1943, 1953; Hart 1932). A sample taken in fall is made up of fish which have finished that year's growth, and the size at age of these fish will be similar to the size at age of fish 1 year older from a sample taken the following spring. This introduces a variability equal to 1 year's growth into the comparisons I have made.

The second source of variation is variation in growth between year-classes. Kennedy (1943) presents data on growth of year-classes from 1926 to 1938 in Lake Opeongo which show considerable variability from year-class to year-class (Fig. 3). Growth of the fish declined from the 1926 year-class to the 1931 year-class, then increased again. Kennedy suggested that the decline in growth rate from 1926 to 1931 was due to recovery of the population from exploitation during the 1914–18 war. However, available information (Fry and Martin personal communication) suggests that this fishing was light, and it seems to me unlikely that the population would still be showing such dramatic effects so many years later. Nor does such an explanation account for the obvious increase in growth for year-classes after 1931. The variations in growth of year-classes shown by the Opeongo fish are extreme (see Edsall 1960), yet they do show that growth can vary considerably in an unexploited population, and even between a mixture of year-classes.

The third source of variation is from errors in aging of fish. Fortunately, most of the problems in aging arise in aging old fish which are normally past the period of most rapid growth.

MORTALITY AND GROWTH

It is important to the understanding of production in unexploited populations to know if there is interaction between mortality and growth. A tentative examination of the hypothesis that populations suffering high mortality also have a high growth rate was significant over all the data ($P < .05$). Most populations exhibiting low mortality came from the southern warmer areas of the whitefish's range, however, where growth was on average slower. Within climatological areas there was no significant difference in growth rate between populations having high and low mortality

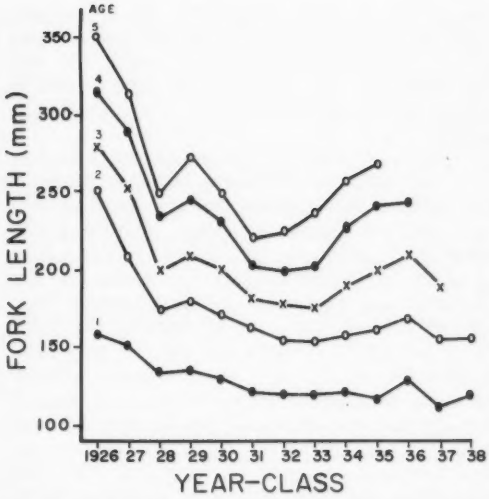

FIG. 3. Average length at age for 13 year-classes of whitefish from Lake Opeongo. Data from Kennedy (1943).

rates. Further study may reveal important environmental parameters limiting growth. For the present, however, the apparent lack of a relationship between mortality and growth will be important in the interpretation of production estimates.

EXPLOITED POPULATIONS

Although it is not possible at this stage to pin down all the factors contributing to variation in growth among unexploited populations, published information indicates that when whitefish are exploited they grow faster (Miller 1947, 1949, 1956; Bond and Turnbull 1973; Schlick 1971d). To explore further the changes in growth rate resulting from exploitation of whitefish I used available information on the growth of whitefish from exploited populations (Table 4) to make two important comparisons: before and after comparisons of growth rate in a population when fishing pressure changed, and comparisons of the growth of whitefish from heavily, moderately, or lightly exploited populations. This latter comparison required a certain amount of subjective judgement in assigning populations to one or another exploitation category. Populations were assigned to exploitation categories as an initial exercise, without reference to growth data. Heavily exploited populations were those in which 50% or more of the regular commercial catch was 4 years old or younger. Five lakes fell into this category: Lake Erie (Van Oosten and Hile 1947), Northern Lake Michigan (Roelofs 1958), Red lakes (Peterka and Smith 1970), Pigeon Lake (Miller 1947, 1956), and William Lake (Schlick 1971f). Some subjectivity was involved in assigning Pigeon Lake and William Lake to the highly exploited category. In 1942 most of the fish in the Pigeon Lake catch were older than 4 years, but this declined until by 1946 most fish were younger than 4 years old. Information from William Lake was sketchy but the catch in test nets was composed substantially of fish 4 years old or younger and annual yield from the lake was about 4 lb/acre (4.5 kg/ha). Lakes subject to moderate fishing pressure had a considerably more youthful age structure than unexploited stocks but not so extreme as the heavily exploited stocks. I judged nine populations to be moderately exploited: south Georgian Bay (Cucin and Regier 1965), Lake Winnipeg (Kennedy 1954), Lake Ontario (Hart 1931a), Wabamun (Miller 1947), Contact and Sulphide (Koshinsky 1965), Morton, Landing, and the Manitoba area of Reindeer Lake (Schlick 1971a, d, e) and Buck Lake (Bidgood 1972, 1973). For Georgian Bay, Lake Winnipeg, Buck, Wabamun, Morton, Landing, and Reindeer lakes the bulk of commercial catches were fish 4, 5, and 6 years old, whereas usually 50% of more of the fish were younger than 6 years old. Thus, these populations appeared less heavily exploited than those assigned to the heavily exploited category. Age structure of samples from commercial catches in Lake Ontario, used by Hart to estimate growth rates, indicated slightly older aged fish, with the bulk of the sample being 7 or 8 years old and with 50% or more less than 8 years old. Judgement about Contact and Sulphide lakes was made in the absence of age structure data and was based on the annual yield of 1.5 lb/acre. This is a rather low yield, and yield alone is not really suitable for assigning exploitation level, as it depends so much on interactions within the biotic community as a whole. However, this yield is high relative to most lakes judged lightly exploited, yet older aged fish were still present in the samples used for age and growth determinations.

Lightly exploited populations were those in which the effect of exploitation on age structure had been relatively slight. Seven lakes fell into this category: Great Slave (Bond and Turnbull 1973); File and Big Sand lakes (Schlick 1971b, c); La Ronge (Rawson and Atton 1953; Qadri 1968); MacKay and Hebden (Koshinsky 1965) and Nipigon (Hart 1931a). In assigning populations

to the lightly exploited category, a great deal of subjective judgement was used. For Great Slave and Nipigon it was fairly clear that exploitation had not had a drastic effect on the fish population. Fish 10–20 years old were still fairly common, and in the case of Great Slave the bulk of the commercial catch was from ages 9 to 12. Rawson and Atton did not provide data on the age structure of the commercial catch in Lac La Ronge, but yield from the fishery for the 10 years preceding their study averaged only 0.1 lb/acre (0.1 kg/ha), indicating very light fishing. Qadri's data show that many old fish were present in the lake. MacKay and Hebden were judged lightly exploited because their annual yield was less than 1 lb/acre (1.1 kg/ha), substantially less in MacKay (0.24 lb/acre; 0.27 kg/ha). File and Big Sand were most difficult of all to categorize. The age structure of catches from them suggested a moderate level of exploitation, whereas yields of whitefish were quite low and suggested light exploitation. Great Slave, La Ronge, and Nipigon were clearly exploited only lightly, therefore, whereas File and Big Sand could be considered light to moderate. MacKay and Hebden are probably lightly exploited.

Mortality rates supported this rather subjective categorization. Those populations classified as heavily exploited, for which mortality estimates were available, had total mortalities from 0.75 to 0.94 with a mean of 0.79. Moderately exploited populations had mortality rates ranging from 0.64 to 0.75 with a mean of 0.68. Lightly exploited populations had total mortality rates ranging from 0.36 to 0.57 with a mean of 0.47, about the same as mortality of unexploited populations.

An increase in growth rate of whitefish following an increase in exploitation rate is documented for three populations (Table 5): Pigeon Lake, Alberta (Miller 1947), Great Slave Lake (Bond and Turnbull 1973), and Landing Lake, Manitoba (Schlick 1971d). Pigeon Lake whitefish grew to be 5 cm larger at age 4 after an increase in fishing pressure that more than doubled yield from the lake, from 7 to 16 lb/acre (7.9–18.1 kg/ha). Young Great Slave Lake whitefish, after a number of years of relatively light exploitation (0.5 lb/acre, 0.56 kg/ha), are now 2–5 cm longer than before exploitation began. The real increase in size of Great Slave fish may be larger since Bond (personal communication) feels they are now aging the fish older than Kennedy (1953) did. A growth increase of Landing Lake whitefish was apparent in samples taken during the 2 years following an unrestricted harvest in which 5 times the usual harvest was taken (Table 5).

Although the growth rate of young fish increased in all three populations, the asymptotic size of fish in Great Slave did not change, even after 25 years of exploitation. Possibly the rate at which the fish grow to asymptotic size is responsive to exploitation, but not maximum size itself.

Growth of whitefish populations after a decline in fishing pressure does not present such a clear-cut picture. Fish from Lake Wabamun showed no apparent change in growth (Miller 1947) after the fishery was closed for 2 years, then gradually reopened. Possibly the closure was too short for any appreciable change to appear. That growth did not change seems peculiar, as, from 1942 to 1946, the average age of the catch shifted 4.4 years to 6.3 years. It seems unlikely that size selection of the net changed, so that in the absence of a change in size at age, the most likely explanation is the appearance of one or two dominant year-classes in the catch. Since the age of the dominant year-class would be about 5 years in 1945, it would have been produced about the time the fishery collapsed in 1940. Its abundance may have precluded any response in fish growth to the closing of the fishery in 1942. Pigeon Lake, on the other hand, showed a clear decline in growth rate after heavy exploitation stopped (Miller 1956).

Comparison of growth rates in relation to degree of exploitation indicates the extent to which whitefish are able to respond to exploitation through increasing their growth. Fish from the five populations classified as heavily exploited grew very rapidly, in all instances approaching the maximum growth displayed by unexploited populations (Fig. 4A). Three of the five heavily exploited lakes (Erie, Michigan, Red lakes) fall in climatic zones having more than 2200 degree days above 42 F (1220 days above 5.6 C) in the growing season, so that the growth of whitefish from these lakes prior to exploitation should have been in the lower range of growth rates for whitefish.

Fish from populations rated as moderately exploited show a much wider scatter of growth curves, yet still have a relatively high rate of growth (Fig. 4B). At age 4 the slowest growing fish from the highly exploited populations were 350 mm long, whereas for the moderately exploited populations the slowest growing fish were only 275 mm long (Fig. 4A, B). Some of the moderately exploited populations grew as quickly as the fastest growing unexploited populations, but none grew faster.

Fish from lightly exploited populations show a still wider scatter of growth curves (Fig. 4C). Two populations (File and Big Sand lakes) grew

TABLE 4. Fork lengths (millimeters) of whitefish from exploited populations.

	Michigan								Erie				S. Ontario			Pigeon		Ontario													
Lake	Big Bay de Noc	Garden Island	Port Aux Basques	South Fox Is.	Gull Island	High Island	St. Helena Island	All sites except High Is.	Sandusky	Loraine	Erie	All Ports	Georgian Bay	Winnipeg	Nipigon			Port Belville	Pleasant	Consecon.	Port Credit	All Ports	Red lakes	Great Slave	Waba-mun	Morton	Big Peter Pond	William	Reindeer	Contact	Sulphide	Landing
Reference	Roelofs 1958								Van Oosten and Hile 1947				Cucin and Regier 1965	Kennedy 1954	Hart 1931a	Miller 1947	Miller 1956	Hart 1931a					Peterka and Smith 1970	Bond and Turnbull 1973	Miller 1947	Schlick 1971a	Rawson 1957	Schlick 1971f	Schlick 1971e	Koshinsky 1965		Schlick 1971d
Age																															
1	140	131	112	98	118	125	154	126	305	326	280	279				104							151		98		142				145	
2	249	227	225	159	199	207	241	217	362	360	335	349			123	208	384					227	283		190		183	325	304	185	213	
3	353	318	330	225	281	291	337	307	400	391	410	418	386	367	168	296	419		227			290	362	197	261	330	224	381	339	236	274	324
4	425	404	414	300	380	386	421	391	423	434	434	446	434	406	194	350	439		290			373	414	250	312	339	262	433	397	277	323	347
5									457	455	–	478	480	420	254		465	368	377		464	451	448	300	340	367	302	487	415	312	358	370
6									478	466	489	506	521	431	269		490	425	434	482	465	462	473	317	368	394	340	496	445	345	381	399
7									493	502	505	530	556	442	281		518	428	469	496	502	508	493	354	387	406	373	504		373	399	437
8									516	–		568	580	451	316			468	515	548	517	499	498	375	402	–	391			399	411	471
9									523	539		568	596	469	361			439		541	540	541		391		496	411			419	422	486
10									530	527		568	609	476	402			543		–		–		406			432			437	432	485
11									546	507		578	609	491	431					–		633		420			450			452	442	509
12									557			602	615	420	441					–				416			475			465	450	521
13									564			610	606		449					633							485			478	457	
14									561			614	623		454																	
15												–	626		488																	
16												648	623		495																	
17															504																	
18															486																	
19															474																	
20															586																	
21															–																	
22															–																	
23															–																	
24															619																	

TABLE 4. Fork lengths (millimeters) of whitefish from exploited populations.

All sites except High Is.	Erie				S. Ontario					Ontario																				
	Sandusky	Loraine	Erie	All Ports	Georgian Bay	Winnipeg	Nipigon	Pigeon		Port Belville	Port Pleasant	Port Consecon.	Port Credit	All Ports	Red lakes	Great Slave	Waba-mun	Morton	Big Peter Pond	William	Reindeer	Contact	Sulphide	Landing	File	Big Sand	La Ronge	MacKay	Hebden
	Van Oosten and Hile 1947				Cucin and Regier 1965	Kennedy 1954	Hart 1931a	Miller 1947	Miller 1956	Hart 1931a					Peterka and Smith 1970	Bond and Turnbull 1973	Miller 1947	Schlick 1971a	Rawson 1957	Schlick 1971f	Schlick 1971e	Koshinsky 1965		Schlick 1971d	Schlick 1971b	Schlick 1971c	Rawson and Atton 1953	Kushinsky 1965	
126	305	326	280	279				104							151		98		142				145						
															283		190		183	325	304	185	213		279		160	157	140
217	362	360	335	349			123	208	384					227	362	197	261	330	224	381	339	236	274	324	304	406	203	211	183
307	400	391	410	418	386	367	168	296	419		227			290	414	250	312	339	262	433	397	277	323	347	339	378	241	259	221
391	423	434	434	446	434	406	194	350	439		290			373	448	300	340	367	302	487	415	312	358	370	378	425	284	297	254
	457	455	–	478	480	420	254		465	368	377		464	451	473	317	368	394	340	496	445	345	381	399	400	454	315	330	284
	478	466	489	506	521	431	269		490	425	434	482	465	462	493	354	387	406	373	504		373	399	437	469	483	345	361	312
	493	502	505	530	556	442	281		518	428	469	496	502	508	498	375	402	–	391			399	411	471		533	368	386	335
	516	–		568	580	451	316			468	515	548	517	499		391		496	411			419	422	486			394	409	361
	523	539		568	596	469	361			439		541	540	541		406			432			437	432	485			417	427	381
	530	527		568	609	476	402			543		–		–		420			450			452	442	509			437	442	401
	546	507		578	609	491	431					–		633		416			475			465	450	521			457	455	419
	557			602	615	420	441					–							485			478	457				477	465	434
	564			610	606		449					633															503	475	452
	561			614	623		454																				526	483	467
				–	626		488																						480
				648	623		495																						493
							504																						
							486																						
							474																						
							586																						
							–																						
							–																						
							619																						

TABLE 3. Fork lengths (millimeters) of whitefish in unexploited populations.

Lake	Long	Grace	Kam	Trout	Michel	Dunnet	Alexie	Baptiste	Chitty	Drygeese	Nueltin	Mac-Donald	MacEwan	Great Bear	Great Slave	Hottah	La Martre	Neso	Payuk	Mink Narrows	Big Athapa-puskow	Little Athapa-puskow	Big Twin	Little Twin	Cliff	Second Musqua-cook	Webster	Trout	Munising Bay	Shake-speare Island	Opeongo	Ge
Reference	Healey and Woodall 1973						Healey unpublished data				Kennedy 1963			Kennedy 1949	Rawson 1949, Kennedy 1953	Wong and Whillans 1973	Bond 1973	Kliewer 1970							Fenderson 1964			Hile and Deason 1934	Edsall 1960	Hart 1932	Kennedy 1943	Gibs Joh 19
Age																																
1	190		170				180		169							129	155	172	161	178	174	189			103	110	92	90		151	161	
2	205		225		190	215		198	202	190						175	197	186		212	244	231	187		156	156	131	131	168	195	183	
3	240	225	375	190			245	209	268	206				254	167	234	264	299	273	274	323	275	305		190	190	167	169	193	224	201	2
4	265	315					245	302	287	270			423	279	210	283	302	367	347	330	380	348	339	369	217	226	205	203	224	259	221	2
5	320	345	440	260	310	335	336	312	337	301			465	292	245	296	375	408	388	387	422	402	414	400	242	251	239	232	236	270	243	3
6	325	375		305	340	385	335	407	391	331			504	373	295	369	429	440	485	445	457	457	460	447	262	285	269	258	249	291	270	4
7	350	405			370	420	415	401	398	367		367	539	411	338	409	441		488	471	495	492	490	491	283	312	287	281	259	321	292	4
8	360	425		430	390	410	430	432	425	461	381	417	557	455	363	434	505								305	341	301	308	270	339	314	4
9	370	440		415	410	435	447	432	413	485	456	433	583	477	385	462	511								316	348	319	340	290	355		4
10		445		450			440	438	428	474	481	452	597	493	402	481	532								330	357	348	358	303	391		
11		460	535	430	455		445	442	413	475	489	469	521	420	491	554									366			374	318	409		
12		470		470			461	430		480	500	481		528	429	499	573								405			384	352	415		
13		475		470	455					450	507	485		551	444	501	590											401	353	417		
14		505		440							533	527		561	456	504												411		445		
15											514			579	466	537														456		
16											536			626	475	537													377	454		
17														635	483	554														467		
18														686	491	561														465		
19														686	498															476		
20														711	505																	
21															510															481		
22															516	562														487		
23															522																	
24																																
25																														506		
26																632																
27																														558		

TABLE 3. Fork lengths (millimeters) of whitefish in unexploited populations.

Alexie	Baptiste	Chitty	Drygeese	Nueltin	Mac-Donald	MacEwan	Great Bear	Great Slave	Hottah	La Martre	Neso	Payuk	Mink Narrows	Big Athapa-puskow	Little Athapa-puskow	Big Twin	Little Twin	Cliff	Second Musqua-cook	Webster	Trout	Munising Bay	Shake-speare Island	Opeongo	George
Healey unpublished data				Kennedy 1963			Kennedy 1949	Rawson 1949, Kennedy 1953	Wong and Whillans 1973	Bond 1973	Kliewer 1970							Fenderson 1964			Hile and Deason 1934	Edsall 1960	Hart 1932	Kennedy 1943	Gibson and Johnson 1969
180		169							129	155	172	161	178	174	189			103	110	92	90		151	161	
	198	202	190						175	197	186		212	244	231	187		156	156	131	131	168	195	183	
245	209	268	206				254	167	234	264	299	273	274	323	275	305		190	190	167	169	193	224	201	219
245	302	287	270			423	279	210	283	302	367	347	330	380	348	339	369	217	226	205	203	224	259	221	284
336	312	337	301			465	292	245	296	375	408	388	387	422	402	414	400	242	251	239	232	236	270	243	359
335	407	391	331			504	373	295	369	429	440	485	445	457	457	460	447	262	285	269	258	249	291	270	409
415	401	398	367		367	539	411	338	409	441		488	471	495	492	490	491	283	312	287	281	259	321	292	433
430	432	425	461	381	417	557	455	363	434	505								305	341	301	308	270	339	314	440
447	432	413	485	456	433	583	477	385	462	511								316	348	319	340	290	355		440
440	438	428	474	481	452	597	493	402	481	532								330	357	348	358	303	391		
445	442	413	475	489	469	521	420	491	554									366			374	318	409		
461	430		480	500	481		528	429	499	573								405			384	352	415		
			450	507	485		551	444	501	590											401	353	417		
				533	527		561	456	504												411		445		
				514			579	466	537														456		
				536			626	475	537													377	454		
							635	483	554														467		
							686	491	561														465		
							686	498															476		
							711	505																	
								510															481		
								516	562														487		
								522																	
																							506		
									632																
																							558		

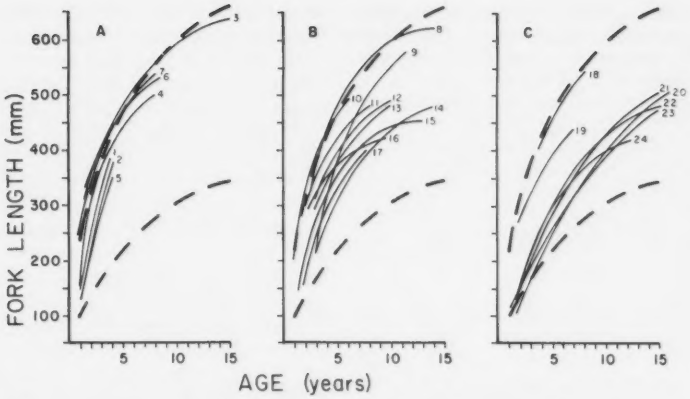

FIG. 4. Growth of whitefish from heavily (A), moderately (B), and lightly (C) exploited lakes. Dashed lines — upper and lower limits of growth in unexploited populations. Lakes are: 1. Michigan, 2. Michigan (High Island), 3. Erie, 4. Red, 5. Pigeon (low), 6. William, 7. Pigeon (high), 8. Georgian Bay, 9. Ontario, 10. Buck, 11. Reindeer, 12. Landing, 13. Winnipeg, 14. Contact, 15. Sulphide, 16. Morton, 17. Wabamun, 18. Big Sand, 19. File, 20. La Ronge, 21. Nipigon, 22. MacKay, 23. Hebden, 24. Great Slave.

quickly. This could merely reflect the high growth rates sometimes observed in northern populations. On the other hand, these were the most heavily exploited of the lightly exploited lakes and may be incorrectly categorized. The remaining populations show growth near the lower limit of growth for unexploited populations in similar climatic zones.

Several conclusions can be drawn from these observations. Clearly whitefish populations respond to exploitation by increasing their growth rate, and it appears as though the rate of response is quite rapid. All populations classified as heavily exploited had growth rates at or near the maximum rate observed for unexploited populations, but did not exceed that rate. This suggests that the maximum growth rates observed may represent a biological maximum which is generally applicable to all whitefish populations. If this is so, then the difference between growth of a population prior to exploitation and this maximum represents the range of response possible in the population, and has considerable relevance to estimates of yield and management techniques.

Reproduction

In examining reproduction in whitefish I have tried to examine the range of values in unexploited populations, and to see how exploitation influences reproduction. Several authors present data on maturity in unexploited or very lightly exploited populations of whitefish (Wong and Whillans 1973; Fenderson 1964; Kennedy 1943, 1949, 1953). In addition I have data on maturity of whitefish from a number of northern lakes. Considerable difficulty exists in assigning stage of maturity to whitefish on the basis of gross examination of the gonads. Authors have used different criteria for the assignment of maturity and the particular method used is not always detailed. However, for most of the data on unexploited populations, it is possible to present the observations as percent nonvirgin fish (the percentage of fish that have spawned at least once, whether or not they will spawn in the year of capture). Assigning small fish to the categories virgin or nonvirgin is not easy, especially in the spring, and is probably subject to considerable interobserver error as well as actual mistakes in classification.

Age of maturity varies widely among unexploited populations. It is difficult to assess how much of this variability is due to errors in assignment to the nonvirgin category or to real differences between populations. The youngest fish classified as nonvirgin were age 3. Only one population, that in Clear Lake, northwestern Maine (Fenderson 1964), showed any appreciable number of nonvirgin fish before age 6. Most populations were close to 100% nonvirgin by age 9 (Table 6).

Because of differences in criteria for assigning maturity, comparison of age or size at maturity

TABLE 5. Change in length at age of whitefish resulting from an increase in exploitation pressure in three lakes. A — before harvest increase, B — after harvest increase. See text for explanation of Landing Lake.

	Lake					
	Pigeon		Great Slave		Landing	
Age	A	B	A	B	B_1	B_2
1	91	103				
2	181	206		170		
3	254	295	167	222		324
4	296	348	210	254	343	351
5	331		245	293	350	378
6	354		295	319	378	419
7	371		338	350	419	448
8			363	373	472	474
9			385	392	485	480
10			402	409	480	518
11			420	418	498	518
12			429	416	521	
13			444	435		
14			456	431		
15			466	452		
16			475	—		
17			483	460		
18			491	505		
19			498			
20			505			

is difficult. However, two authors (Edsall 1960; Fenderson 1964) present data on four populations in which maturity was assigned in the same way, and I have data on nine populations near Yellowknife in the Northwest Territories which may be compared with Edsall's and Fenderson's results (Table 7). Results from the nine Northwest Territories populations must be treated with caution because sample sizes were small. Nevertheless, the results indicate age to maturity ranging from 3 to 10 years and sizes ranging from 204 to 400 mm fork length. In general the Northwest Territories populations for which I have data matured at a larger size, but not necessarily an older age, than the relatively more slowly growing southern populations.

These observations on unexploited populations may be compared with the available data for exploited populations. Miller (1947, 1956) reports that before fishing pressure was increased on Pigeon Lake, only a few fish spawned when 3 years old, but most were mature as 4-year-olds. After fishing pressure increased, all of the 2-year-old fish became mature, indicating a dramatic decline in age to maturity associated with the increased fishing pressure. Average fork length of 2-year-old whitefish in 1946 was 236 mm, whereas the average length of 3-year-old fish in 1939 was 250 mm and of 4-year-old fish was 296 mm, suggesting that the fish matured at a smaller size as well as at a younger age.

A similar decline in age of maturity is seen in the Great Slave Lake fishery over the years. Kennedy (1953) recorded whitefish as maturing first at 5 years of age and 226 mm fork length during the early years of the fishery. It was not until they were 9 years old and 358 mm long that more than half the fish were nonvirgin. Recent data (Bond and Turnbull 1973) show that the fish now mature first when 4 years old and 254 mm fork length, and that half are mature at age 6 when they average 320 mm fork length. Again there is a slight suggestion of reduced size at maturity associated with the reduction in age at maturity. If Bond is aging fish older on average than did Kennedy, changes in age at maturity would be even greated than indicated.

Among heavily exploited populations, data on age at maturity are available only from Pigeon Lake and Lake Erie (Van Oosten and Hile 1947). As previously mentioned, Pigeon Lake fish are mature at age 2. Considerable numbers of Lake Erie fish were mature at age 2, but full maturity was not reached until age 4. This was in spite of the fact that Lake Erie fish were as large or larger than Pigeon Lake fish of the same age. South Georgian Bay whitefish, an example of a moderately exploited stock (Cucin and Regier 1965), began maturing at age 3 but never appeared to be fully mature. Again differences in methodology influence the results. Cucin and Regier (1965) recorded as mature those fish from which sexual products could be expressed. The highest percentage mature by this technique was 83% at age 4. Probably 100% of these fish would have been recorded as mature if the gonads had been examined. Four-year-old Georgian Bay whitefish averaged about 435 mm fork length, slightly larger than Lake Erie fish of the same age.

Limited though they are, the available observations on age and size at maturity suggest that if mortality is increased through exploitation, whitefish mature at a younger age and possibly also at a smaller size.

In applying data on reproduction to problems of production and yield, one is really concerned with recruitment. If recruitment is reduced under heavy exploitation, then without sufficient growth compensation, yield will decline. One possible cause of lowered recruitment is reduced egg deposition or reduced brood stock in the population.

TABLE 6. Percent nonvirgin fish at various ages in unexploited whitefish stocks.

Lake	Source	Age 1	2	3	4	5	6	7	8	9	10	11	12
Hottah	Wong and Whillans 1973	0	0	18	7	17	79	94	96	100	100	100	100
Clear	Fenderson 1964		0	20	30	100	100	100	100	100	100	100	
Cliff	Fenderson 1964							80	90	100	100		
Musquacook	Fenderson 1964								65	70	100		
Opeongo	Kennedy 1943		0	10	?	?	?	90	100	100	100		
Great Bear	Kennedy 1949						0	10	58	97	100		
Great Slave	Kennedy 1953			0	0	2	5	12	30	59	78	91	98
Combined Y.K. lakes	Healey unpublished data	0	0	0	18	30	69	85	96	100	100	100	100

TABLE 7. Age and size at first maturity in 13 unexploited whitefish populations.

Population	Reference	Age (*yr*) ♂♂	♀♀	Size (fork length *mm*) ♂♂	♀♀
Munising Bay, Lake Superior	Edsall 1960	7	10	260	272
Clear	Fenderson 1964	3	4	227	
Cliff		3		204	
Second Musquacook		5		272	
Alexie	Healey unpublished data	5	6	350	350
Baptist		6	7	350	350
Chitty		6	7	350	350
Drygeese		8	8	400	400
Kam		3	3	340	360
Grace		6	5	325	350
Long		6	6	340	320
Michel		8	7	385	355
Trout		8	8	395	395

Miller (1956) explored the possibility that brood stock was reduced as one explanation for the failure of the Pigeon Lake fishery. When decline in age at maturity was taken into account, there was no relationship between year-class strength and apparent size of the brood stock in the populations. Considering that the average size of fish was larger after heavy exploitation, it seems reasonable that if the size of the brood stock was not diminished, then egg deposition was not diminished either. Christie (1968), however, presents estimates of egg deposition for Lake Ontario whitefish which indicates a rather striking decline over the 14-year period preceding 1956. At the same time, virtual population estimates of year-class strength were the same or higher, indicating an increased survival of spawn so that recruitment was not adversely affected. It appears possible, then, that heavy fishing might reduce brood stock to the extent that egg deposition would be affected. How this would affect recruitment is uncertain.

Reproductive responses to exploitation can involve reduced age or size at maturity. They can also involve increased fecundity (Nikolsky 1962). Data on fecundity are very few for lake whitefish but what is available refers to heavily (Lawler 1961), moderately (Cucin and Regier 1965; Hart 1931b; Christie 1963), and lightly (Kennedy 1953; Qadri 1968) exploited stocks as well as unexploited stocks (Healey and Nicol 1975). Average fecundity for the heavily exploited Lake Erie stock was 16,300 eggs/lb; for the moderately exploited Georgian Bay and Lake Ontario stocks it was 8200 eggs/lb and 9900 eggs/lb, respectively; for the lightly exploited Great Slave and La Ronge populations it was 6753 eggs/lb and around 8000 eggs/lb, respectively. Average fecun-

dity of unexploited Northwest Territories stocks for which I have data ranged from 9017 eggs/lb to 12,227 eggs/lb. There appears, therefore, to be no relationship between fecundity and rate of exploitation.

Population Size

There is no satisfactory information on changes in population size of whitefish resulting from exploitation. This information is vital, but at present can only be guessed at in a general way. One way is to consider virtual population estimates for fisheries which exploitation pressure has changed. Davidoff et al. (1973), for example, present virtual population estimates for many years of fishing on Lake Winnipeg during a period of continuously increasing fishing mortality. For the period 1940–46 when total annual mortality ranged from 0.59 to 0.72 with a mean of 0.67, virtual population estimates averaged 563,000 fish. For the period 1956–61 when total annual mortality was higher (0.83 to 0.90, mean 0.86 for 1959 to 1961), virtual population averaged 529,000 fish. The earlier set of estimates involved mainly ages 5 and older with median age fluctuating around 6 years, whereas the latter set of data involved mainly fish aged 4 and older with median age between 4 and 5 years. This means that total population was probably significantly smaller in later years, yet the vulnerable portion of the stock was of similar size.

For Pigeon Lake Miller (1956) provided virtual population estimates which covered the years of heavy exploitation and some years following the reduction in heavy fishing. A number of poor brood years during the period of heavy fishing contributed to a low average virtual population of 75,700 fish for the year-classes 1938–44, all of which contributed to the period of heavy fishing. The average virtual population for year-classes 1945–48, which were exploited during the years of lower effort, was higher at 103,700 fish. However, it might be argued that the small 1944 year-class should also have been included in this average as it also contributed during a number of years of low fishing. This would make the average year-class strength during years of low fishing 85,208, or much nearer to that in the years of high exploitation. As with the Lake Winnipeg data, however, the mean age of fish in the catch was considerably younger during the years of heavy fishing. Although the vulnerable population was probably similar at the two fishing pressures, total population was almost certainly lower during the period of heavy fishing.

Another way of approaching the question of changes in population size with change in exploitation is to examine catch-per-unit-effort in relation to effort. Peterka and Smith (1970) present catch and effort data for Red lakes. Their data indicate a fairly constant effort of between 3860 and 8086 units (mean 6114 units) from 1930 to 1941. Effort increased in 1942, and from 1942 to 1951 ranged between 8454 units and 14,816 units (mean 11,549 units), so that effort was almost twice as great during the latter period. If the vulnerable population declined during this period of heavy fishing, then catch-per-unit-effort should have been less. However, catch-per-unit-effort averaged 1.77 lb from 1930 to 1941, and 2.71 lb from 1942 to 1951. The catch-per-effort figures, in fact, suggest an increased vulnerable population.

It would be of considerable value to know with some certainty how much total population declines with fishing. However, if the vulnerable population remains the same over a fairly wide range of efforts, then changes in growth will be the determining factor in maintaining yield. If growth improves enough to prevent a decline in average weight, then yield will remain high. It was increases in growth with fishing which kept yields high in the Pigeon Lake fishery (Miller 1947, 1956). Growth appears to have been unable to keep pace with declining age of capture in Lake Winnipeg (Davidoff et al. 1973) and in Lake Ontario (Christie 1963), so that yields declined in these fisheries as exploitation became more intense. The alternatives of increasing or decreasing vulnerable populations with increased fishing cannot be ruled out at this stage. The results of Peterka and Smith (1970) suggest an increased vulnerable population with increased fishing, whereas some of Christie's (1963) results suggest a decline in the vulnerable population in Lake Ontario, possibly because of reduced spawn deposition.

Yields in Relation to Fishing Effort

Provided recruitment is not interfered with, a stable yield can be achieved at virtually any level of total mortality. What is important in a fishery is to maintain as high a yield as possible consistent with whatever other social and environmental constraints are pertinent. It is worth examining at this point yields from fisheries at different extremes of exploitation to see what light this sheds on the problem of high sustained yields from whitefish populations. The data I gathered are not precise and in some instances simplistic as-

TABLE 8. Yield of whitefish in lb/acre from lakes suffering different fishing intensities.

Fishing intensity					
Low		Moderate		High	
Lake	Yield	Lake	Yield	Lake	Yield
Churchill	0.51	Winnipeg	0.48	Red	0.36
Big Peter Pond	4.64	Georgian Bay	0.36	Pigeon	17.00
Ile à la Crosse	0.48	Wabamun	6.00	Erie	0.47
La Rouge	0.10	Wintering	0.87	William	4.10
Great Slave	0.50	Landing	3.00	Winnipeg	0.12
Big Sand	0.53				
File	0.64				

sumptions had to be made in estimating yield per unit area. It is yield per unit area which is important, as the fisheries were located on lakes of different sizes. Yields in pounds per acre were estimated for five lakes suffering high exploitation: Pigeon (Miller 1947), Red lakes (Peterka and Smith 1970), Erie (Van Oosten and Hile 1947), Winnipeg recent data (Davidoff et al. 1973), and William (Schlick 1971f). Estimated yield per acre is probably reasonable for Pigeon, Red lakes, and William. I estimated yield from Lake Erie from landing statistics in Van Oosten and Hile (1947) and the area of the U.S. portion of Erie. Yield from Winnipeg I estimated from catch data and the estimated area of the north basin of Winnipeg. Apart from Pigeon, which was unable continuously to support such a high yield, none of the heavily exploited lakes had a spectacular yield (Table 8). Red lakes, Erie, and Winnipeg, in fact, had small yields per acre. Of moderately exploited lakes, six provided data on yield per acre: Winnipeg (Kennedy 1954), Georgian Bay (Cucin and Regier 1965), Wabamun and Pigeon (Miller 1947; Bidgood 1972), and Wintering and Landing lakes (Schlick 1971d, g). Again, apart from Wabamun, Pigeon, and Landing lakes, yields were less than 1 lb/acre (Table 8). Seven lightly fished lakes, Churchill, Big Peter Pond, Ile à la Crosse (Rawson 1957), La Ronge (Rawson and Atton 1953), Great Slave (Bond and Turnbull 1973), and File and Big Sand (Schlick 1971 b, c) also had yields less than 1 lb/acre except Big Peter Pond. Yield is apparently extremely variable regardless of what level of exploitation is applied. This is understandable, considering the wide range of biotic and abiotic features occurring in the lakes considered, in particular the range of fish fauna present. Nevertheless, even the most extreme levels of exploitation did not produce continuing yields lower than those commonly observed in lightly exploited lakes, and some were substantially higher. The same is true for the moderately exploited lakes. Although with such variable data it is not possible to demonstrate any real improvement in yield at higher fishing intensities, the information does not indicate a decline in yield at high levels of fishing.

Biomass and Production in Whitefish

My purpose in beginning this manuscript was to consider the dynamics of exploited whitefish populations with a view to improving management technique. Because of variability among whitefish populations, I propose not to discuss particular examples of natural populations, but to discuss representative situations of growth and mortality. The approach I shall take is to consider the distribution of biomass and production in representative whitefish populations and how such populations would respond to exploitation.

Growth and mortality are so variable that the first step must be to examine how these parameters influence the distribution of biomass and production in populations. Growth rate (as opposed to growth pattern) obviously will influence the total amount of biomass and production but will not influence the distribution of biomass and production among the age-classes. Mortality rate, on the other hand, strongly influences the distribution of both production and biomass. As mortality increases, the peak of biomass and production is shifted to the youngest ages. At 0.20 annual mortality, for example, peak biomass occurs at age 7, at 0.40 mortality the peak occurs at age 3; and at 0.80 mortality the peak occurs between age 1 and 2 (Fig. 5). As mortality increases, the curves of biomass and production become more and more skewed, with more and more of total biomass or production concentrated in the first years of life. Production is not shown in Fig. 5, but it follows the same pattern as biomass, only

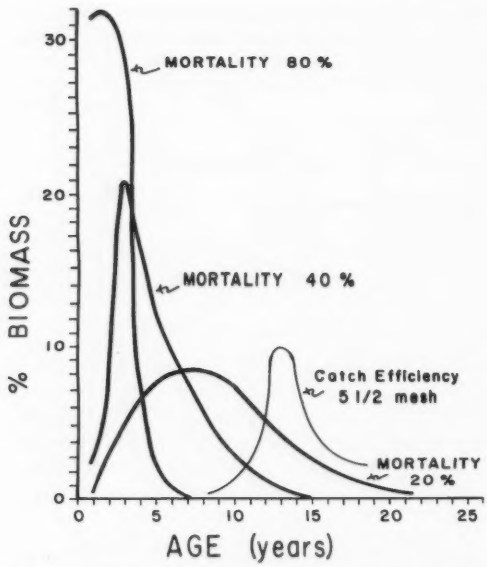

FIG. 5. Distribution of biomass in populations suffering different rates of mortality.

the curves are skewed more to the left. The curves in Fig. 5 are based on the assumption that mortality is constant from age 1 to the oldest ages. If mortality is less for intermediate age-groups, then the lower the average mortality, the older the age of maximum biomass. However, the general conclusions about exploitation and the response of whitefish populations will not change.

According to fishery management theory, best yields are obtained if the population is cropped at the age of maximum biomass. For practical purposes it is impossible to apply such a rule to many whitefish populations. For most populations, the age of maximum biomass is an age at which individual fish are not of marketable size. However, it is worth examining in some detail what is likely to happen when one begins to harvest populations such as those represented in Fig. 5.

An approximate harvest efficiency curve for 5½-inch mesh, gillnetting is superimposed on the biomass curves of Fig. 5. It is immediately obvious that a considerable percentage of the population suffering only 20% mortality is accessible to the 5½-inch mesh, and only an infinitesimal proportion of the biomass of the population suffering 80% mortality is available to the same net. When exploitation begins on the population with a natural mortality rate of 20%, a considerable capital stock of fish is available to the fishery, and the fishery can, in the course of its development, have a considerable influence on the total stock of fish. If the lake is large, the capital stock available to the fishery will also be large, and it will take some time for effects of the fishery to be felt. However, it is inevitable that eventually the fishery will significantly reduce total biomass, opening the way for compensatory responses on the part of the population, such as increased growth. Progress of the fishery should then be that increased growth rate will cause the net efficiency curve to shift toward the younger ages. The exact point at which maximum yield will be achieved will depend upon the interplay of changing growth rate, changing mortality rate, and changing stock size. Such a fishery, depending on the size of the lake, would be characterized by quite a number of years in the "fishing up" state during which the fishery would expand slowly as it cropped the large accumulated stock. Eventually, however, the fishery would reduce this stock to the point where compensation would begin to occur, and the fish would grow faster. As growth increased, successively younger ages would become available to the fishery, and these, because of their relative abundance, would tend to hold yields high. Eventually, however, the population would reach the point where further growth compensation was impossible. Yield and catch-per-unit-effort would then decline as the population finally came into equilibrium with its new total mortality rate. The exact time span of this process is difficult to predict. It would be influenced by size of the lake, initial intensity of the fishery, changes in intensity, changes in gear and efficiency, relative availability of other species, and so on. For example, Lake Erie appears to have reached the last stages in this development process in the 1940s (Van Oosten and Hile 1947), Lake Ontario not until the 1950s (Christie 1963), and Lake Winnipeg not until the 1960s (Davidoff et al. 1973). All of these lakes have long exploitation histories beginning in the 1800s and have experienced considerable changes in gear, techniques, and fishing intensity over the years (e.g. Hewson 1960; Regier and Loftus 1972).

When a fishery begins on a population suffering a high natural mortality, however, the story is somewhat different. Only a small fraction of total biomass is initially available to the fishery. Although the initial progress of the fishery will be similar to that of the population with low mortality, the stage of fishing up will be relatively shorter, catch-per-unit-effort will be smaller, and

the total impact of the fishery on the population will be quite small. The impact may be small enough to elicit only a nominal compensation on the part of the whitefish population, so that equilibrium will be approached without much, if any, progression down through the age-classes. In such a situation the population will approach equilibrium, with the fishery still dependent on the older unproductive age-groups. Some fisheries appear to have developed in this way. Recent developments on Great Slave suggest that this may be what is happening to this fishery.

Growth, Mortality, Exploitation, and Yield

If it is possible to generalize from the information presented on whitefish biology, then certain features of populations should provide a clue to fishery potential and the best way to harvest the population. Assuming that the difference between growth of the unexploited population and maximum growth observed represents the potential of the fish to respond to exploitation, then populations with the best fishery potential would be those which have a low natural mortality and a slow growth rate. Such a population can absorb a high fishing mortality and has ample scope for compensation through increased growth. Also, the combination of low mortality and low growth rate would suggest a relatively dense population with few natural enemies. In such a situation competition between fishing and other sources of mortality would be low, and the relatively high population density and capacity for compensation through increased growth would indicate a relatively high equilibrium yield. Because of the initial distribution of biomass, such a population could be exploited with relatively large mesh nets throughout its history. However, it should be recognized that initial catches taken from the capital stock of the population will be high, and one should expect a considerably lower catch-per-unit-effort when the population finally comes into equilibrium.

The situation in which the unexploited population has a high mortality and a low growth rate appears to have reasonable fishery potential as well. The low growth rate suggests a high population density, although most of the population biomass will be concentrated in the younger age-classes. Probable yields from such a population will be lower than in the previous situation because of the competition between fishing and natural mortality. Nevertheless, the population has good scope for compensation because of its initial growth rate. Such a population might have to be harvested in a slightly different manner, however, than the population with low mortality. Because the larger older fish form such a small proportion of population biomass, harvesting with large mesh nets may not reduce the population enough to elicit compensation. The controlled use of smaller mesh nets may be required to force the population into a high growth rate. Because of the limited stock of large fish available, initial catches will not be so dramatic as in the population with low natural mortality, and the decline in catch-per-unit-effort will not be so dramatic as equilibrium is approached.

A situation with limited fishery potential would be the situation in which mortality was low, but growth was high. Such a situation could result from a limited spawning habitat, or very high egg and larval mortality, followed by low mortality after age 1. In such circumstances population density is low and growth rate is high. Although there would be little competition between fishing and natural mortality, recruitment is limited by some factor not related to fishing, and because of the high natural growth rate there is little scope for compensation in the population. Such a population should be harvested only lightly with large mesh nets, and care should be taken that the brood stock is not significantly reduced. If it is known that reproduction is limited by some physical constraint such as a shortage of suitable spawning habitat, then a reduction in brood stock would not be serious. However, if recruitment is limited by heavy mortality of eggs and larvae, reduction in brood stock could lead to depensatory mortality (Neave 1953).

Probably the least satisfactory situation from a fishery point of view is when the unexploited population has both a high mortality rate and a high growth rate. Competition between natural and fishing mortality and the lack of any scope for compensation in the population would preclude anything but a very small yield from such a population.

Changes in growth rate provide whitefish with a very powerful compensating mechanism (Healey 1973). It appears that in most circumstances, best yield from the stock would come when exploitation was heavy and the fishery depended upon very young age-classes. However, achievement of this situation has not generally led to high sustained yields. In fact, as fisheries approached this point yields have generally fallen (Christie 1963; Davidoff et al. 1973). This could simply be a result of approaching equilibrium and the absence of younger ages to support a fic-

titiously high yield as outlined above. Or it could mean that exploitation had exceeded the ability of the population to compensate through increased growth. In addition, populations having a very youthful age structure appear to be very susceptible to violent oscillations (Lawler 1965; Christie 1963). Occasional brood failure or above average survival are not damped out over a number of years, but are almost immediately reflected in yields. Also, when the fishery depends principally on fish aged 2, 3, and 4, most fish get only one chance to spawn, and many never spawn at all. One or two years of brood failure can severely reduce recruitment to the fishery and brood stock in subsequent years. So, although there appear to be theoretical reasons for supposing that best yields would be obtained with heavy exploitation, this situation is relatively unstable and collapse of the fishery in such situations is probable. From the point of view of stability, it would be best to maintain the fishery at a more moderate level of exploitation, so that ages 5, 6, and 7 contributed most to the fishery. Such an age structure would mean that most fish would have the chance to spawn at least once, many would spawn twice, and some even 3 or 4 times. This additional breeding potential would mitigate against violent oscillations in year-class strength through brood failure, and would introduce the further damping effect of several years of life before each year-class entered the fishery. Although real maximum yields might be missed in a few instances, the increase in stability would more than compensate.

The statements above should be generally true, although they would have to be qualified to conform to specific instances. The advantage to using these observations for management purposes is that populations do not have to be treated according to some rule of thumb, but can be examined on their own merits. Ways in which our knowledge of whitefish biology could be used to help assess unexploited populations have been outlined above. It is possible by the same technique to assess a population at any stage in its exploitation history. The relevant information would be age structure, growth rate, and proportion mature at each age. Such information is easily obtained from survey sampling and in some instances from sampling the commercial catch. These three pieces of information will allow the manager to relate growth to the probable range for the climatic area in which the lake occurs, and to the probable maximum growth rate for the fish, thus giving him a measure of the scope for compensation left in the population. It will allow him to estimate total mortality rate which, together with the age structure of the catch, will tell him how close the population is approaching to optimum conditions of stability and yield. Data on maturity will tell him if the population is responding appropriately to exploitation through reduced age to maturity, or if the brood stock is being threatened by exploitation. In short he can gauge the health and resilience of the stock and decide whether it can withstand continued exploitation at the same level, or whether exploitation should be increased or decreased. Various observed values of these parameters and how they can be interpreted to gauge the stage of exploitation of the population are outlined in Table 9. The table provides a number of clues to making decisions about fishery management which are consistent with the biology of the fish. Such decisions will, of course, be tempered by local conditions and individual experience.

As an exercise in how the system might be applied I have analyzed the Great Slave Lake fishery. As I pointed out in the introduction, catches in the fishery have recently fallen off and this necessitates a reevaluation of its management. The data necessary to evaluate the fishery according to the scheme outlined in Table 9 are included in Bond and Turnbull (1973). Whitefish growth rate, in spite of some increase over the years, is still relatively low, indicating considerable scope for compensation. Mortality is moderate, so that the capability is there to absorb increased fishing mortality. In fact, fishing mortality has been negligible over the years, as shown by a comparison of the present mortality rate and that found earlier by Kennedy (1953). Because mortality caused by large mesh nets is uneven, the fishery has reduced the abundance of the oldest year-classes, but has not materially affected the age-classes which make up the bulk of the catch. The age structure is still relatively old, even in the more heavily fished areas, indicating that numerous year-classes are present that do not contribute to the fishery. Maturation has responded to fishing and the fish now mature at a younger age, so there need be no immediate fear of jeopardizing the brood stock. Because of the moderate mortality rate, most of the biomass and production in the population is concentrated in the youngest age-classes. The cumulative percent biomass and production in the population are shown in Fig. 6. Percentage biomass and production are summed from the oldest ages back to the youngest in this figure so that fish aged 1 year and older contain 100% of total production and biomass; fish aged 2 and older contain about 92% of total production and biomass; fish aged 3 and

TABLE 9. Interpretation of growth, mortality, age structure, and maturation in management of whitefish populations.

Growth

1. Low: Considerable scope for response to increased fishing. Indicates underexploitation.
2. Moderate: Some scope for response to increased fishing. Indicates moderate exploitation.
3. High: Little scope for response to increased fisning. Indicates heavy exploitation.

Mortality

1. Low: Considerable scope to absorb increased fishing. Considerable biomass present in the older ages and available to the fishery. Indicates either a young fishery or underexploitation.
2. Moderate: Some scope to absorb increased fishing. Less biomass available in older ages. Fishing might be increased if other factors are favorable but reduced mesh size might be required to get an improved yield.
3. High: Little scope to absorb increased fishing. Biomass concentrated in younger ages. Indicates heavy exploitation.

Age structure

1. Old: Numerous unexploited year classes which could contribute to the fishery, fishing could probably increase.
2. Moderate: Some unexploited year classes. Fishing might be increased if other factors favorable.
3. Young: Few or no unexploited year classes. Particularly unstable situation. Fishing should probably be reduced a bit.

Maturation

1. Fish responding to fishing by maturing at a younger age. Fishery probably healthy.
2. Fish not responding to fishery by maturing at a younger age. Fishery may be in danger.

older contain about 37% of total production and 82% of total biomass, and so on. These curves show nicely the infinitesimal proportion of production and biomass in the older age-classes where fishing is concentrated. Estimated catch efficiency curves for both 4½- and 5½-inch mesh gillnets are superimposed on the curves of production and biomass. Both fall well away from the major concentration of biomass and production in the population. From these curves, one can estimate the percent of total production or biomass which is available to 4½- and 5½-inch mesh. For 5½-inch mesh, the present minimum mesh, only 0.252% of production and 1.17% of biomass are available to the fishery. There is a sustainable yield from this portion of the population, but it is very small, probably less than 0.1% of total production in the population. The situation if 4½-inch mesh were permitted is much better, for 1.337% of production and 4.14% of biomass are available to the 4½-inch mesh, more than 3 times the biomass and 5 times the production. Even if no compensatory growth occurred with the introduction of smaller meshes, the probable yield would be much better than the equilibrium yield with 5½-inch mesh.

The recent declines in catch suggest that increases in fishing gear efficiency are no longer able to bolster yield from an unproductive portion of the stock, and that the fishery is coming into equilibrium while still dependent on the older unproductive age-classes. The use of 5½-inch mesh in this instance has not been sufficient to elicit much compensation on the part of the fish. This is understandable, considering the tiny proportion of the stock which was available to this mesh. My evaluation would be that the stock can support a more intensive fishery, and that there is good reason to consider the introduction of smaller mesh to achieve a better harvest. Although the average size of fish in the catch would be somewhat lower if 4½-inch mesh were introduced, the increase in catch would more than make up for it.

This overview of the Great Slave fishery is purposely simplistic to illustrate application of the hypotheses I have presented. The whitefish population in Great Slave is not homogeneous, nor is

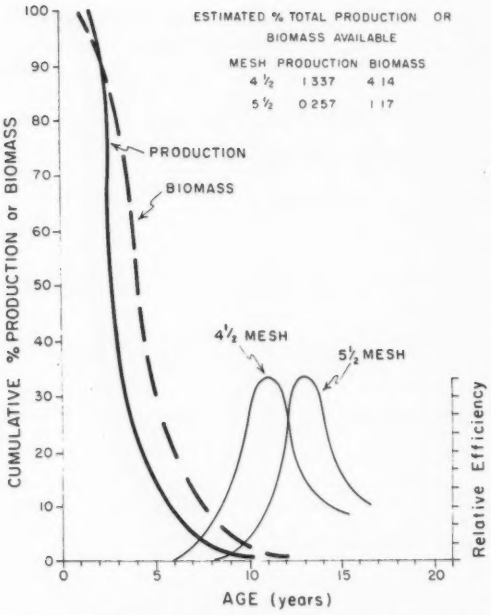

FIG. 6. Cumulative percent distribution of biomass and production in the Great Slave Lake whitefish population. See text for further explanation.

fishing pressure evenly applied. The areas around Hay River and Moraine Bay are much more heavily fished than in other areas. However, the same techniques may be applied to the management of subpopulations or statistical areas. No change in philosophy is required as most large lakes are presently managed by statistical areas of some sort.

Conclusions

The features of biology of whitefish presented in this manuscript suggest that there are simple ways of assessing the capacity of an unexploited population to support a fishery, and of assessing the status of exploited populations in terms of probable maximum yields. The method does not depend upon the assignment of a particular yield which the population should produce. This is an impossible task, because the biotic and abiotic features differ so much from lake to lake that a common maximum yield seems out of the question. Instead the method proposed is to seek a balance between fishing and natural mortality such that yield will be maximized and stable. Theoretical considerations indicate that yield will be maximized at high levels of exploitation in many populations, but that population structure is unstable at these levels. A compromise situation that permits high yield yet good stability appears to be more desirable.

The parameters upon which an assessment of the capacity of a whitefish population to support a fishery can be based are simple parameters of growth, age structure, and maturity, which can readily be gained from survey sampling of the fish stock or sampling in the commercial fishery. The principal advantage of such assessment is that it uses the fish as an integrator of all the interacting facets of the community, and lets the fish tell what the state of the system is. As we are generally ignorant of the functional relationship between fish production and such parameters as competition, food supply, and habitat, using parameters of the fish population itself as an index of productivity is probably the least risky approach to management.

Acknowledgments

A major stimulus for this review was discussions with Freshwater Institute management biologists about problems of managing northern whitefish stocks. Preliminary drafts and the formulation of ideas owe much to the patient criticism of my colleagues at the Institute, especially Dr K. Patalas. Dr W. A. Kennedy of the Nanaimo Pacific Biological Station and Dr T. G. Northcote of The University of British Columbia also read and criticized the manuscript.

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The Management of Marine Fisheries. *J. A. Gulland. Bristol: Scientechnica (Publishers) Ltd., Rome, 1974. 184 p., $19.80.*

The management of marine fisheries can be said to have reached a stage best described as the end of the beginning. That traditional fisheries are limited biologically in the degree to which they can be exploited is now widely accepted, and the implications of this have been important in recognizing that major changes in the concepts of ownership and jurisdiction are required, if management is to operate effectively within these biological limits. In the northwest Atlantic, a system of internationally negotiated management is being elaborated and tested; by its very success at the level of agreement and acceptance of convention and principle, it is uncovering major problems of application and the corresponding complexity of the management mechanisms that are needed to solve them.

John Gulland's book on marine fisheries management is therefore timely. It is difficult to think of many other individuals with the same breadth of experience and depth of involvement in the management initiatives of the last two decades. His views would be worth serious study on this ground alone. In fact, what he does succeed in doing is to create a sense of reality; we are brought down to earth. Anyone who has regularly attended meetings of international agencies which have been struggling to solve the overfishing problem — and through which most of the recent advances have been made — should recognize the management scene as described and discussed by Gulland. Gulland's picture, far more than the standard texts on the application of ecological theory to management problems or the volumes of symposia papers on management represents what actually happens. It is easy enough to repeat the platitude that management of fisheries is a complex mixture of biological, economic, social, and political problems; it is quite another thing to demonstrate how these factors have interacted in specific cases and with what results — and this is what Gulland has done.

One has learned to expect lucid thought and clear, even trenchant expression from Gulland and with a few minor lapses this is what one gets. He prefers the complexities of the problem to become apparent as the reader follows two important case histories of management in separate chapters at the beginning of the book. That of the Atlantic whale fisheries is an admirably clear exposition of a subject that attracts great public interest, although not everyone would go as far as Gulland in his final assessment — that rational management is now within reach. Recent correspondents to Nature express different views. The North Atlantic trawl fisheries present far more complex problems, both biologically and in the number and variety of competing interests. Here, of course, the pace of events is so rapid that it is impossible for the author of a book to be completely up to date and the impact of the most recent developments of the last few years is not covered, although it is anticipated.

Gulland did not intend to produce a text book. It is in any case far too early to produce a text book on marine fisheries management. However, in tracing briefly the history of the fisheries themselves and the development and operation of bodies such as ICES and ICNAF, and indeed, throughout the book, Gulland, on almost every page, produces a statement, opinion, or implication that is of more general significance and importance. One can instruct oneself by making a list of these as one reads on, each item deserving further study and amplification.

In discussing the biological aspects of management, Gulland stresses the basic types of assessment models that have proved capable of providing the relevant advice, but with the minimum of mathematics. The models themselves are simple in principle and these principles can and should be thoroughly understood by anyone with an involvement in management decisions or an interest in their impact. Equally deserving of study and thought are Gulland's prefatory statements that the basic requirement of the biologist's prediction should be that it is commensurate with the uncertainties in the social, political, and economic fields, and that the purpose of a model is to organize the available information so as to facilitate the best choice between alternate management strategies. Although the models currently being used would be made more precise and effective by an improvement in the statistical and sampling data on which they depend for their validity and precision, for the most part they are adequate.

A thoughtful reading of Gulland's chapter on management objectives will indicate why this is

so. In his discussion of the broadly economic factors in relation to management objectives it is reassuring to the fisheries biologist to find how easily currently acceptable economic models can be incorporated and assimilated in his own stock in trade, but it is also evident that specific economic (and even more so, social and political) objectives will not be as easily identified and achieved as biological objectives. What is required is an information base and a mechanism for translating this information into advice and decision. It has taken many years to reach even a reasonable standard for fisheries biology as Gulland demonstrates; it does not seem likely that the transition from general theory to specific practice in the less precise and therefore more difficult areas will come overnight.

Gulland's treatment covers a period when most major initiatives were being pursued through the agency of international regional fisheries bodies, and in a chapter on the mechanics of international fishery management he pulls together, in a convenient form, some very useful information on these bodies. No less than 22 are tabulated and described. Even so it has not been possible to deal with the most recent developments in the most successful of these — ICNAF — which by now has limited total catches for 64 individual stocks, each with national allocation of shares, and has introduced a "two-tiered" quota system as an approach to the by-catch problem together with a number of other management techniques, whose effectiveness it is too early to judge. Incidentally, the amendment to Article VIII of the ICNAF Convention mentioned by Gulland on page 111 as being "in the process of ratification" and which substitutes "optimum utilization" for "maximum sustainable yield" (MSY) as a specific objective of the work of the Commission was in fact ratified by December 1971 and acted on almost immediately (in January 1972) in connection with the setting of herring total catch limitation and the allocation of national shares.

With the prospect of increased national jurisdiction and with Canada one of the important countries exercising this jurisdiction and responsibility, we must be aware of a point made early on by Gulland and also in the final chapter on the future of management of fisheries. The national concentration of jurisdiction and its use to satisfy national aspirations will inevitably internalize problems that have had to be faced and solved at the international level. It is easy to assume that a 200-mile economic zone, or whatever emerges from the Law of the Sea Conference, will eliminate national problems. This is not so and those in Canada with the responsibility for dealing with the problems that have already emerged as a result of decisions at the international level will, I am sure, heartily concur. There is, however, no reason to suppose that the basic nature of the problems will be radically changed or that knowledge and experience gained during the present stage of management are not relevant. It is for this reason that Gulland's expressed intention in writing this book — in the hope that in some small part it would lead to an understanding of the problems of fisheries management — should be realized. That he hopes also that it will result in the authorities concerned being able to ensure a better use of fish resources remains a hope; to which we can all say Amen!

This book is the first of a series of monographs on Marine Science by a new publishing firm, and one hopes that later ones will be as interesting and stimulating and in a similarly convenient format. That so much has been gotten into less than 200 relatively small pages is not the least impressive aspect of this book.

However, it would have benefited from one more editing operation, for there are some noticeable defects. Some of the two-part figures should have been labelled A and B (as others are) and referred to in the text separately; a line of type has been omitted in a citation at the top of page 104; from pages 98 to 101, figures are wrongly referred to (surely it should be Fig. 17 and not Fig. 16 on page 99), do not have units listed on the axes, and have minor grammatical errors or misprints which make this important section (on stock and recruitment) more difficult to follow than it should be. The Index, too, is not particularly well done; "Permanent Commission" is mentioned twice in the text (pages 56 and 62) but referred to only once in the Index. This example is chosen because the term, in any case, is not defined in the text.

This should not be allowed to detract from its value as one of the most realistically informative books on the subject. It is, also, good value for money. Anyone who claims to be a fisheries manager, in whatever role, should read it, and anyone at all interested in marine affairs would enjoy reading it.

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(R20)

COMING NEXT MONTH/À PARAÎTRE LE MOIS PROCHAIN

Life histories and abundance of crustacean zooplankton in the outlet of Lake Superior, 1971–72
J. H. Selgeby

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S. N. Messieh

Analysis of tri-aryl phosphate esters and the determination of Imol S-140 in fish tissue and water samples by gas chromatography
D. A. J. Murray

Incidence of trematode parasites of American plaice (*Hippoglossoides platessoides*) of the Scotian Shelf and Gulf of St. Lawrence in relation to fish length and food
J. S. Scott

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D. S. Cherry, K. L. Dickson, and J. Cairns Jr.

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W. Kwain

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Translation Series

The following translations have been prepared by or for the Fisheries and Marine Service, for use in its investigations. A limited number of copies are available for distribution as long as the supply lasts, or may be obtained by interlibrary loan, from the station where the translation has been deposited. Information on translations generally may be obtained from the Office of the Editor.

Traductions

Les traductions dont il est question ci-après ont été préparées pour ou par le Service des pêches et des sciences de la mer, à des fins de recherche. On peut en obtenir un nombre limité d'exemplaires ou se les procurer par prêts entre bibliothèques, aux stations dépositaires de ces textes. On peut en général se renseigner sur ces traductions au Bureau du rédacteur.

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Journal of the Fisheries Research Board of Canada

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Content

We invite submission of manuscripts in the broad field of fisheries management, ocean science, and aquatic environments relevant to Canada.

Articles: Reports of research studies of broad scope that are original contributions to science.

Notes: Differ from papers on the basis of scope or completeness, rather than quality of research. They may be on problems with narrow, readily defined limits; on work that is largely confirmatory; on methodology or research techniques; on advances in knowledge arising as by-products of broader studies; on progress in projects that were discontinued; or on preliminary findings that warrant publication before problems are resolved. They may be organized with as much flexibility as articles except that they have no centered headings and should occupy no more than four printed pages.

Reviews: Detailed articles that are critical appraisals of broad areas of investigation, particularly in fields that have shown rapid development in recent years.

Perspectives: Essays of opinion or hypothesis on fisheries, ocean science, aquatic environments, or related topics that are of concern to the professional and layman. References are desirable; informal style is welcomed.

Letters: Opinion on topics recently under discussion in the *Journal*; preferred maximum length, 500 words. The Editor reserves the right to shorten letters and to make other changes to accord with the style of the *Journal*.

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The *Style Manual for Biological Journals* (3rd ed. American Institute of Biological Sciences, 3900 Wisconsin Ave. NW., Washington, D.C.) is used as the authority in matters of form. The standards for spelling are: *Webster's Third New International Dictionary* and *le Grand Larousse Encyclopédique*. Titles of periodicals are abbreviated as in the *Biological Abstracts* list. References to literature are made by the name-and-year system. The *Journal* follows the names and spelling recommended in *A List of Common and Scientific Names of Fishes from the United States and Canada* (Third Edition, 1970) Special Publication No. 6 of the American Fisheries Society.

Series titles should be avoided. An *abstract* is required for all manuscripts, and should state concisely, in less than 250 words, what was done, the essentials of the new knowledge, and the main conclusions. Abstracts are published in English and French; those submitted in one language only are translated by the *Journal*.

The Editor normally submits each manuscript to at least two *referees*. Contributors are encouraged to suggest at least four names to be added to the list from which referees may be selected.

Illustrations should be submitted in triplicate as photographs (*glossy prints*) no larger than a standard page and must accompany the manuscript. Prints should not be mounted, stapled, or clipped. Symbols, letters, and numerals must be neat, and at least 1.5 mm high when the illustration is reduced to the appropriate size. Drawings should, when possible, be grouped to facilitate comparisons.

Proofs are sent to the author, who is responsible for correcting only errors in typesetting. *Alterations to the content must be avoided.*

Fifty *reprints* are supplied free. Additional reprints may be purchased at the time of publication (an order form is sent out with the galley proof). All orders should be sent to the Printing Department, University of Toronto Press, 5201 Dufferin Street, Downsview, Ont. M3H 5T8. Invoices relating to the reprints will be sent out by the Printer. A charge of $30.00 per page is made for articles published. When submitting a manuscript, an author should state that his institution will accept this charge.

Checklist on Manuscripts

Attention to the following questions will expedite appraisal of manuscripts by referees and editors.

Are the findings, interpretations, and conclusions adequately documented and relevant to the purpose of the study?

Are all the tables and figures needed, and organized to facilitate comparisons? Are there inconsistencies between tables or figures and the text, or within the text? Should some of the data be made available separately, in a manuscript report or at a data repository? If you use statistical analysis are the statistics subordinate to the research?

Would any of the text be clearer if condensed? Are summary statements given at the beginning of sections and paragraphs, and are details in sections and paragraphs relevant to their topics? Does the organization of the manuscript follow logically from the statement of purpose in the introduction?

Does the *title* encompass the content of the report?

Does the *abstract* give the essentials of the new knowledge?

Is the *introduction* largely limited to the scope, purpose, and rationale of the study? Is review of the literature limited to defining the problem?

Are details of *materials and methods* limited to what scientists need in understanding the design of the study and in judging adequacy of the data?

Are generalizations on the *results* adequate? Are findings distinguished from inferences from them?

Is *discussion* limited to interpretation and significance of the findings?

Journal de l'office des recherches sur les pêcheries du Canada

Guide des Auteurs

On peut se procurer au bureau du Directeur de la publication un guide plus détaillé pour la préparation de manuscrits à partir de thèses.

Sujets

Nous désirons reçevoir des manuscrits s'insérant dans le vaste domaine de la gestion des pêches, des sciences de la mer et des milieux aquatiques au Canada.

Articles: Comptes rendus d'études d'une portée générale, constituant des contributions inédites à la science.

Notes: Se distinguent des articles par la portée et le degré d'achèvement plutôt que par la qualité des recherches. Elles peuvent traiter de problèmes particuliers et bien définis; de travaux qui sont en grande partie confirmatoires; de méthodologie ou de techniques de recherche; de progrès de la connaissance découlant d'études plus vastes; de développements dans des projets abandonnés; ou de découvertes préliminaires qui méritent d'être publiées avant la solution de problèmes à l'étude. Les notes peuvent être présentées d'une façon aussi souple que les articles, mais elles ne doivent pas porter d'en-têtes centrés ni occuper plus de quatre pages.

Critiques: Articles détaillés constituant une appréciation critique de vastes champs de recherches, particulièrement dans les domaines ayant connu de rapides progrès au cours des dernières années.

Perspectives: Essais de l'opinion ou de l'hypothèse sur les pêches, les sciences de la mer, les milieux aquatiques, ou sur des sujets connexes qui intéressent professionnels et profanes. Il est préférable de donner des références et d'écrire, si possible, dans un style simple.

Lettres: Opinions sur des sujets exposés dans le *Journal*. Longueur maximale préférable: 500 mots. Le directeur se réserve le droit de raccourcir les lettres et de faire d'autres modifications dans le style du *Journal*.

Dispositions générales

Le *Style Manual for Biological Journals* (3e éd., publié par l'American Institute of Biological Sciences, 3900 avenue Wisconsin N.O., Washington, D.C.) sert de guide quant à la forme de présentation. Le *Webster's Third New International Dictionary* et le *Grand Larousse Encyclopédique* font autorité pour l'orthographe des mots. Les titres de périodiques sont abrégés suivant les *Biological Abstracts*. Les références bibliographiques dans le texte doivent suivre le système nom de l'auteur-année. Pour les noms de poissons et leur orthographe, le *Journal* suit les recommandations contenues dans *A List of Common and Scientific Names of Fishes from the United States and Canada* (Troisième édition, 1970), Publication spéciale No. 6 de l'American Fisheries Society.

Il faut éviter les *articles en série*. Un *résumé* doit accompagner chaque texte et doit exposer brièvement (moins de 250 mots) le travail accompli, les points essentiels des connaissances acquises et les principales conclusions. Les résumés paraissent en anglais et en français, le *Journal* veillant à la traduction dans le cas des présentations unilingues.

Le directeur soumet d'ordinaire chaque texte à l'approbation de deux *arbitres* ou plus. Nous encourageons les collaborateurs à suggérer au moins quatre personnes dont les noms pourraient figurer sur la liste des arbitres.

Les *illustrations* doivent être sous forme de photos (*sur papier glacé*), ne pas dépasser les dimensions d'une page normale et accompagner le texte. Elles ne doivent pas être montées, coupées ni brochées. Les symboles, lettres ou chiffres doivent se détacher avec précision et avoir au moins 1,5 mm de hauter lorsque l'illustration est réduite à la grandeur voulue. Les dessins doivent, autant que possible, être groupés pour faciliter l'étude comparative.

L'auteur doit revoir les *épreuves* qu'on lui remet et en corriger seulement les erreurs typographiques. *Il doit éviter de changer le contenu du texte.*

Cinquante *tirés à part* sont fournis gratuitement. Les tirés à part peuvent être achetés au moment de la publication. Un bon de commande à cet effet accompagne les épreuves; il doit être retourné en même temps que celles-ci, non pas au bureau de la rédaction, mais au Printing Department, University of Toronto Press, 5201 Dufferin Street, Downsview, Ont. M3H 5T8, qui se chargera de la facturer. Les frais sont de $30.00 par page pour la publication des articles. Quand un auteur soumet un manuscrit, il doit spécifier que son institution accepte les frais.

Listes de contrôles pour manuscrits

Une attention particulière aux questions suivantes facilitera l'évaluation des manuscrits par les arbitres et les éditeurs.

Les découvertes, interprétations et conclusions sont-elles bien documentées et pertinentes au but de l'étude?

Les tableaux et figures sont-ils tous nécessaires et agencés pour faciliter les comparaisons? Y a-t-il contradiction entre les tableaux ou les figures et le texte, ou dans le texte même? Est-ce que certaines données devraient être présentées séparément, dans un rapport manuscrit ou dans un dépôt de données? Si vous faites une analyse statistique, la statistique cède-t-elle le pas à la biologie?

Le texte y gagnerait-il en clarté s'il était plus condensé, en tout ou en partie? Contient-il des exposés sommaires au début de chaque section ou paragraphe, et les détails de ces sections et paragraphs son-ils pertinents au sujet traité? La marche du manuscrit est-elle conforme à la logique énoncée dans l'introduction?

Le *titre* est-il concis, tout en étant représentatif du contenu du rapport?

Le *résumé* donne-t-il l'essentiel des connaissances nouvelles?

L'*introduction* se limite-t-elle à une esquisse de la portée, du but et des raisons de l'étude? La revue de la littérature se limite-t-elle à la définition du problème?

La section *matériel et méthode* contient-elle le minimum de détails qui permettront aux autres scientifiques de saisir les grandes lignes de l'étude et de juger de la valeur des données?

Les généralisations sur les *résultats* sont-elles adéquates? Permettent-elles de distinguer entre constatations et déductions?

La *discussion* est-elle limitée à l'interprétation et à l'évaluation des découvertes?

Journal of the Fisheries Research Board of Canada

Guide for Typists Preparing Copy

Read "Guide to Authors" in the current volume of the *Journal*. When in doubt about how to type or arrange details, follow *Journal* practice.

Type the manuscript on white bond paper, not flimsy, preferably size 21.5 × 28 cm (8.5 × 11 inches). If possible, use paper with the lines numbered in the margin. Leave all margins at least 2.5 cm wide.

Double-space all parts of the manuscript, including tables, captions for illustrations, footnotes, and the reference list. Underline only material that is to be printed in italics. Indent the first line of each paragraph in the text and of each footnote.

In titles and headings, capitalize only the first word and proper names and adjectives. For small capitals, use small letters.

Indicate all text footnotes by superscript arabic numerals. Number them consecutively throughout the manuscript, starting with those on the title page. Place each footnote immediately below the reference to it, separating it from the text by horizontal lines.

On the first page type only the title, the authors' names and affiliations, related footnotes, and the running head (see Appendix A). Type the abstract on a separate page (see Appendix B). Number all pages, beginning with the title page and including those for tables and captions for illustrations.

Have the author check the finished manuscript, and send the ribbon copy and two other clear copies to the Office of the Editor.

Tables: Examine the current volume of the *Journal* to see how tables are set up (see Appendix C).

Note especially the following concerning *numbered tables:*

Type each on a separate page, using paper without the lines numbered.

Number them with arabic numerals.

Use horizontal lines above and below the headings and below the columns, and seldom elsewhere. Never use vertical lines; leave extra spaces instead.

Indicate table footnotes by superscript small letters and type them below the table.

Place the tables after the list of references.

Where each table is first referred to, type in the margin "Table — near here."

Note that *text tables* are not numbered, are typed with the text, and seldom need horizontal lines (see Appendix C).

Illustrations: Send at least 3 *glossy prints* (no larger than 21.5 × 28 cm) of each illustration with the manuscript. Type the author's name and figure number on a strip of paper and fasten it to the back of the print. Do not write or type on the print itself. Also, type the captions in succession on a separate page, headed "Captions for illustrations." Put this page at the end of the manuscript.

Where each figure is first referred to, type in the margin "Fig. — near here."

Mathematical Expressions, Equations, and Formulas: Leave three spaces above and below lines that have much material in superscripts, and above and below equations and formulas. In the various expressions, place each component in its correct position (see Appendix D).

Show all symbols clearly. In the margin, spell out Greek letters the first time they occur, and distinguish between the small letter *l* and the numeral *one*, and between the capital letter *O* and the numeral *zero*. Make other explanatory notes similarly.

References: Check the text to see that all references in the list are referred to. Type the reference list, double-spaced, in the style used in the *Journal* (see Appendix E). Names of journals are abbreviated as in the *Biological Abstracts* list; if an abbreviation is in doubt, type the name in full.

Spellings: About 100 words common in fisheries journals are listed in Appendix F.

Appendices: Appendices A to F are available on request from the Office of the Editor.

Journal de l'office des recherches sur les pêcheries du Canada

Guide pour les secrétaires d'auteurs

Lisez le "Guide des Auteurs" reproduit dans l'édition courante du *Journal*. Si vous avez des doutes sur la façon de dactylographier le texte ou d'en agencer les détails, suivez la pratique du *Journal*.

Dactylographiez le manuscrit sur papier blanc de bonne qualité, et non sur papier pelure, mesurant de préférence 21,5 × 28 cm (8,5 × 11 pouces), laissant toujours une marge d'au moins 2.5 cm. Si possible, employez un papier avec lignes numérotées en marge.

Tout le manuscrit, y compris tableaux, légendes de figures, renvois et références bibliographiques, devra être dactylographié à double interligne. Ne soulignez que les mots qui devront être imprimés en italique. Mettez en retrait la première ligne de chaque paragraphe du texte et de chaque renvoi.

Dans les titres et les en-têtes, n'employez la majuscule que pour le premier mot et les noms propres. Pour les petites majuscules, employez les miniscules du dactylographe.

Indiquez tous les renvois du texte par des chiffres arabes suscrits. Numérotez les renvois consécutivement par tout le texte, à commencer par ceux de la page titre. Placez chaque renvoi directement sous sa référence, le séparant du texte par des lignes horizontales.

La première page ne doit contenir que le titre de la publication, ne nom et l'affiliation des auteurs, les renvois appropriés et le titre courant (voir Appendice A). Dactylographiez le résumé sur une page séparée (voir Appendice B). Numérotez chaque page, à commencer par la page titre, et y compris celles qui contiennent les tableaux et la légende des figures.

Voyez à ce que l'auteur vérifie le manuscrit final, et soumettez la première copie et ainsi que deux copies lisibles au bureau du Directeur de la publication.

Tableaux. Consultez une copie du *Journal* pour la composition des tableaux (voir Appendice C).

Portez une attention particulière à ce qui suit au sujet des *tableaux numérotés*.

Dactylographiez chaque tableau sur feuille séparée, vous servant de papier sans lignes numérotées.

Numérotez les tableaux en chiffres arabes.

Tracez des lignes horizontales au-dessus et au-dessous des en-têtes et au-dessous des colonnes, rarement ailleurs; ne tracez jamais de lignes verticales; laissez plutôt des espaces supplémentaires.

Indiquez les renvois dans les tableaux par les minuscules suscrites et placez les renvois au bas du tableau.

Placez les tableaux à la suite des références bibliographiques.

A l'endroit où le tableau est mentionné pour la première fois, indiquez dans la marge "Tableau — près d'ici."

Remarquez que les *tableaux dans le texte* ne sont pas numérotés, sont dactylographiés avec le texte et ne requièrent que rarement des lignes horizontales (voir Appendice C).

Figures: Envoyez avec le manuscrit trois photos *sur papier glacé* (21,5 × 28 cm ou moins) pour chaque illustration. Dactylographiez le nom de l'auteur et le numéro de la figure sur une lisière de papier que vous collerez au dos. N'écrivez pas à la main ou à la machine sur la photographie même. En plus, dactylographiez les légendes, dans l'ordre, sur feuille séparée, intitulée "Légendes des figures." Intercalez cette feuille à la suite du texte.

A l'endroit où chaque figure est mentionnée pour la première fois, inscrivez en marge "Fig. — près d'ici."

Equations, formules et expressions mathématiques: Laissez trois espaces au-dessus et au-dessous des lignes renfermant des chiffres ou des lettres suscrits et souscrits, ou encore au-dessus et au-dessous d'équations et de formules. Placez chaque composante des différentes expressions dans la position qui lui est propre (voir Appendice D).

Indiquez clairement tous les symboles. Epelez en marge les lettres grecques, la première fois qu'elles apparaissent. Faites une distinction entre la lettre *l* et le chiffre *un*, et entre la majuscule *O* et le chiffre *zéro*. Insérez de la même façon toute autre note explicative.

Références: Vérifiez le texte afin de vous assurer que toutes les références apparaissant dans la liste bibliographique sont bien mentionnées dans le texte. Dactylographiez à double interligne dans le style en usage dans le *Journal* (voir Appendice E). Les noms de revues sont abrégés suivants les *Biological Abstracts*; en cas de doute, écrivez le nom au long.

Orthographe: Une liste d'environ 100 mots communément employés dans les périodiques sur les pêches est donnée en Appendice F.

Appendices: On peut se procurer une copie des Appendices A à F sur demande au bureau du Directeur de la publication.

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JFRBAK32(3)333–455(1975)

Volume 32, No. 3, March 1975

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